APPENDIX B – EXHIBIT 1

The Biological Status in Bonnie Creek, Galum Creek, and White Walnut Creek Following Stream Diversion and Reconstruction

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EXECUTIVE SUMMARY

Comparisons of the fish and benthic macroinvertebrate communities from reconstructed portions of Bonnie Creek, Galum Creek, and White Walnut Creek (Perry County, Illinois) were compared to benthic and fish samples from upstream undisturbed sites in each stream, and a selected non-impacted stream site from Eagle Creek Basin surveyed by IEPA (Matson and Hite 1988). Existing data generated for Bonnie Creek, Galum Creek and White Walnut Creek was obtained from field surveys required as conditions of permits granted to Consolidation Coal Company (Perry County, IL) for diversion and subsequent permanent relocation of portions of these streams in the late 1980's up through 2000. All streams are first or second order small streams from geographical regions that currently include agricultural land uses and previously coal mining activities. The purpose of the comparisons were to determine the status of the biological communities in the reconstructed portions of each stream and assess whether the reconstruction stream reaches supported benthic and fish communities that were equivalent to site pre-relocation or regional reference stream conditions. A regional reference site typical of relatively undisturbed conditions situated within only an agricultural land use watershed could not be found with IEPA data for both fish and macroinvertebrate sufficient for comparison. The reference site within the Eagle Creek Basin is considered conservative for comparison to sites within an agricultural setting as IEPA classified this reference site as non-impacted.

The fish community evaluation was based on the fish IBI following Simon and Dufour (1998) using abundance based data from the sites. The macroinvertebrate community evaluation included comparison of the EPT richness and total richness metrics for spatial and temporal patterns because some of the available data was not abundance data and presented as species presence/absence data. In addition, the macroinvertebrate Index of Biotic Integrity (MIBI) following Plafkin et al. (1989) was used for available abundance data to assess overall status among sites.

Evaluation of the fish data indicated the assemblage at all stream sites exhibited species common to small streams of Illinois. The streams were characterized by a dominance of green, bluegill, and longear sunfish, along with frequent capture of red shiner and sand shiner (Bonnie Creek) and/or blackstripe and bluntnose minnow (Galum Creek and White Walnut Creek). A fish IBI evaluation could not be conducted for White Walnut Creek because presence/absence data only was available. Fish IBI values for all streams evaluated, including the reference stream, were in the low range of values and indicated communities dominated by tolerant species that were primarily omnivores and capable of exploiting a variety of physical habitats.

The benthic macroinvertebrate assemblage at all sites included species common to small streams in Illinois and represented most of the major groups of aquatic insects as well as snails, aquatic isopods and amphipods, flat worms, mussels and clams. The overall abundance of benthic macroinvertebrate organisms in the samples collected was lower than expected at all sites in each stream and the entire sample was likely evaluated to obtain the available data. Confounding factors at the time of sample collection included lack of measureable flow at many sites. However, low specimen counts were common to all sites and collection dates and are not considered a significant factor in interpretation of results. The richness-based metrics for all stream sites were highly variable and comparisons indicated a general, but not conclusive, trend for higher EPT richness in April samples compared to samples collected in August. In contrast, the total richness metric showed a general, but not conclusive, trend for higher richness in August than present in April.

Key findings from the evaluation of fish and benthic macroinvertebrate samples from reconstructed portions of streams with comparison to upstream or regional reference conditions included:

1. Fish IBI values for Galum Creek indicate the reconstructed reach to support a fish community typical of a relatively undisturbed stream within Illinois, and difference in fish IBI value among the undisturbed upstream reach and the reconstructed site was not of sufficient magnitude to indicate a meaningful spatial difference in fish community health or integrity.

2. Fish IBI values for Bonnie Creek in 1997 indicated fish community health and integrity in the reconstructed reach was equivalent to the fish community health and integrity of the undisturbed upstream site BCA. The comparison to Bonnie Creek fish IBI results would also indicate the reconstructed reach of Bonnie Creek supports a fish community typical of a relatively undisturbed stream within Illinois. Bonnie Creek is a tributary of Galum Creek.

3. The benthic macroinvertebrate samples from Galum Creek collected during in August indicates the reconstructed reach attained a macroinvertebrate community equivalent with pre-construction conditions (based on MIBI values) and the Galum Creek macroinvertebrate assemblage was equivalent to the regional reference site. The data indicated recovery of the macroinvertebrate community in Galum Creek occurred within a 5-year time span.

4. Macroinvertebrate samples collected from the reconstructed portion of Bonnie Creek in 1997 were considered equivalent to the upstream benthic community. The benthic macroinvertebrate MIBI values from Bonnie Creek were less than determined for the regional reference site suggesting lower biological integrity. However, based on the relatively consistent results for total richness and EPT richness at both the undisturbed and relocated sites in Bonnie Creek, an MIBI score lower than the regional reference site may be normal for Bonnie Creek.

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1 Introduction

The purpose of this study is to report on the evaluation of fish and macroinvertebrate community recovery following diversion and permanent relocation. The study involved three different permanently relocated streams located in Perry County, Illinois, USA, each of which had available fish and macroinvertebrate data from a relocated stream reach and undisturbed upstream reach. A determination of recovery was based on comparisons of the biological community structure and composition between the relocated stream reach and upstream undisturbed reach, as well as comparison to a regional reference stream. The regional reference stream was selected to represent a site with fish and macroinvertebrate data that was located within a coal mining region of Illinois and was considered undisturbed based on Illinois Environmental Protection Agency (IEPA) biomonitoring results. The goal of this study was to demonstrate recovery of the biological communities within the relocated stream reaches to be structurally equivalent with upstream conditions and/or comparable to an appropriate regional reference condition.

This evaluation used existing data generated for Bonnie Creek, Galum Creek and White Walnut Creek from field surveys required by permits granted to Consolidation Coal Company (Perry County, IL) for diversion and subsequent permanent relocation of portions of these streams in the late 1980's up through 2000. Galum Creek benthic macroinvertebrate assemblage and fish census data were available for 1988 (Pike and Owen 1988), 1993 (Pike and Owen 1993), 1997 (Owen and Pike 1998), 2003 (PEC 2003) and 2006 (PEC 2006). Bonnie Creek benthic macroinvertebrate assemblage and fish survey data were available for 1997 (Owen and Pike 19998), 2003 (PEC 2003), and 2006 (PEC 2006). Benthic macroinvertebrate assemblage and fish survey data were available for White Walnut Creek in 2003 (PEC 2003). Each of these streams is located in Perry County, IL and can be associated with potential impacts from coal mining.

The regional reference stream reflecting minimal impact was selected from among several watershed studies reported by the Illinois Environmental Protection Agency (IEPA). The selected stream site was chosen from the Intensive Survey of the Eagle Creek Basin, Saline and Gallatin Counties, Illinois (Matson and Hite 1987). The Eagle Creek Basin is in extreme southeast Illinois and represents a small second order Illinois stream with minimal reported impacts from mining or significant anthropological activities. This Eagle Creek Basin area and watershed was selected because, like Perry County, historical active or inactive coal mine operations were known to occur within the watershed and that IEPA had conducted intensive surveys of streams within watershed. Biological survey data from site ATE-06 in the Eagle Creek Basin was specifically targeted for comparison to Galum Creek, Bonnie Creek, and White Walnut Creek primarily because the watershed drainage size for this site best matched the drainage area for the relocated stream reaches. A comparable watershed drainage size among all the streams evaluated facilitates a comparison of the fish census data, and likely subjects the biotic communities to similar hydrological patterns. Conveniently, results of the IEPA intensive survey for site ATE-06 also indicated to IEPA that this site likely representative of background conditions for the Eagle Creek Basin Watershed area by exhibiting none of the traditional mining related impacts. The ATE-06 site was characterized by low metals, sulfate, chloride and cyanide concentrations and IEPA stated this site to have no indications of other major water quality impacts (Matson and Hite 1987).

2 Methods

The general approach of this project was to compare standard bioassessment results for sites within the relocated stream reached from Bonnie Creek, Galum Creek, and White Walnut Creek, to bioassessment results for undisturbed sites upstream of the relocated stream reaches, and to bioassessment results for site ATE-06 in the Eagle Creek Basin. Detailed specifics regarding field sample collection efforts, sorting, taxonomic classification and enumeration protocols, and development of metric values and metrics scores were not always sufficient to rely on a direct comparison of conclusions presented in each report.

Bioassessment protocols and the choice of metrics have progressed and improved over the 13-year period of study for the various streams reflected in the monitoring reports, and a direct comparison of bioassessment indices would likely not be based on equivalent measures and result in misleading conclusions. Thus, recalculation of bioassessment metrics and indices using a suite of community composition metrics common to all samples was necessary to alleviate potential problems associated with comparisons of survey results based on a mix of different bioassessment measures or techniques. The recalculation of bioassessment metrics and indices used in this study included generally accepted methods with considerations given to the taxonomic precision of the available data.

Fish bioassessment protocols followed protocols presented in Simon and Dufour (1998) for the Eastern Corn Belt Plain of Indiana applicable to fish survey data from streams with >20 square miles drainage area and less than 1,000 square miles. Assumptions that were made to address certain metric scoring criteria in lieu of missing information included an equal sampling efficiency (catch per unit effort) for all stations and no deformities, eroded fins, lesions, and tumors (DELTS) for all fish captured. Benthic macroinvertebrate bioassessment protocols followed Protocol III of Plafkin et al. (1989) with comparison to the Eagle Creek Basin site ATE-06 as the reference community. It was assumed that similar methods and adequate sampling effort was conducted among all the sites evaluated.

3 Results

3.1 Fish

3.1.1 Galum Creek Fish

Galum Creek fish surveys were conducted at sites GLA (upstream), GLC (relocated reach), and GLD (downstream) during April and August of 1988, 1993, 1997, 2003 and 2006, with the exception of survey dates during May of 1993 and September of 2006. Only presence/absence data were recorded (no enumeration data) for the 2003 and 2006 surveys. Site GLC from 1988 to 1997, and sites GLC2, GLC3 and GLC4 in 2003 and 2006 represented the diversion and relocated portion of Galum Creek. Site GLA was an undisturbed site upstream from the diversion, and site GLD was downstream of the reconstructed stream reach. Flow in Galum Creek was variable from year to year but patterns that existed indicated higher flows in spring than in fall (often no flow in August at the sampling sites) and flow showed a trend to increase from GLA downstream to GLD. Maximum flow during the sampling periods in spring was 30.7 cubic feet per second (cfs) with median flow around 12 cfs compared to a maximum flow of 3 cfs in the fall with the median flow, when present, at 1 cfs or less. Conductivity ranged from 395 to 4868 umhos/cm, and was generally lower during April than during August when flow existed at sites during both sampling periods. The fish assemblage for the three sites were dominated by bluegill sunfish, green sunfish, and longear sunfish, with bluntnose minnows being characteristic at the upstream GLA site; blackstripe topminnow being more common at GLC than other sites; and shiners, especially the red shiner (*Cyprinella*) being characteristic at the downstream site GLD. Flow conditions likely influenced habitat conditions and the presence or absence of species during sampling events.

3.1.2 Bonnie Creek Fish

Bonnie Creek fish surveys that provided abundance data included sites BCA (upstream), BCC (relocated stream reach), and BCD (downstream) during April and August of 1997. Survey results provided presence/absence data only in 2003 and 2006 for sites BCB2, BCB3 and BCB4 that represented the relocated reach of Bonnie Creek; site BCA represented the undisturbed upstream site; and BCD and BG represented undisturbed downstream locations. Flow at the time of the 1997 survey was less than 20 cfs and was higher in April than reported during August (zero flow reported at BCA and BCC). The fish assemblage at all three sites was dominated by shiners (red shiner and sand shiner) and sunfish (green sunfish, bluegill sunfish, and longear sunfish). The upstream site (BCA) exhibited a more diverse and evenly distributed array of sunfish than either of the two downstream sites (BCC and BCD) and a high abundance of the bluntnose minnow, which was rare or absent at the downstream sites. A total of 15 different fish species were reported from Bonnie Creek in 1997 compared to a total of 25 different fish species present in 2006 indicating a increase in the number of resident species in this stream. Within the reconstructed reach the fish species increased from 10 species in 1997 to 18 different species in 2006.

3.1.3 White Walnut Creek

White Walnut Creek fish surveys were conducted at sites WWA (upstream), WWC2 (relocated reach), and WWD (downstream)) in April and August of 2003. Flow at the time of the surveys were extremely low and ranged from zero or too low to measure during August to 0.84 cfs upstream increasing downstream to 3.4 cfs at WWD. Fish data were reported as present/absent data only and a total of 17 different species were listed for White Walnut Creek. Three species (golden shiner, green sunfish, and bluegill) were present at the upstream site WWA while 11 different fish species were reported from the reconstructed reach WWC2, and 10 different fish species were reported from the downstream site WWD. The fish assemblage at WWC2 and WWD included several sunfish, shiners, and minnows. Species only observed at the reconstructed site WWC2 included spotted bass, redear sunfish, orange spotted sunfish, and bluntnose minnow. Species observed only at WWD included the gizzard shad, sand shiner, and tadpole madtom.

3.1.4 Eagle Creek Basin Site ATE-06

The Eagle Creek Basin fish survey was conducted on May 28-29, 1986 by IEPA. Site ATE-06 is located in Upper Eagle Creek and at the time of the fish survey flow was noted to be low. Flow conditions at ATE-06 were presumed to be comparable to the relative flow conditions at the reconstructed stream reaches during fish survey events. A total of seven different fish were collected from site ATE-06 of which four was represented by sunfish species. However, the grass pickerel (*Esox americanus vermiculatus*) was the most abundant fish captured.

3.2 Fish IBI

Fish IBI values could be calculated for Bonnie Creek in 1997 data, Galum Creek for 1988, 1993, and 1997 data, and the selected reference site ATE-06 in the Eagle Creek Basin for the 1986 data. Fish IBI results for Bonnie Creek in 2003 and 2006; Galum Creek for 2003 and 2006, and White Walnut Creek for 2003 cannot be determined because of the presence/absence type of data presented for each stream. Fish community health and quality increases with IBI value. Evaluation of the fish IBI values will focus on relative differences and patterns between the reconstructed reach of the stream to the undisturbed upstream site (seasonal and spatial); comparisons IBI value with the reference site ATE-06 and considered not to be impacted by mining and other land uses; and for Galum Creek the change in IBI value over the multiple-year monitoring period (temporal trends). Fish IBI metric values, metric scores, and the calculated fish IBI value following Simon and Dufour (1998) at site ATE-06, and sites in Bonnie Creek and Galum Creek for April is presented in Table 1, and for August samples in Table 2.

3.2.1 Galum Creek Fish IBI

The fish IBI values for Galum Creek ranged from a low of 22 during August of 1993 at the undisturbed upstream site GLA to a high of 38 at the downstream site GLC during August of 1997. However, fish IBI values at all three monitoring sites were generally the lower during August 1993 and generally higher during August of 1997. No consistent pattern could be identified that indicated a seasonal trend for generally higher or lower fish IBI values during either April or August. Fish IBI values for site GLC within the reconstructed reach of Galum Creek tended to be slightly lower than the undisturbed upstream IBI values from GLA, but this was not consistent. For example, Table 1 shows fish IBI values from GLC were higher than upstream at GLC during April of 1998 (value of 30 and 24, respectively) and equal at a value of 22 during August 1993 (Table 2). Fish IBI values at the downstream site GLD were typically greater than fish IBI values in the reconstructed reach (GLC) at all times. The difference in fish IBI value among the three sites was not of sufficient magnitude to indicate a meaningful spatial difference in fish community health or integrity in Galum Creek. Similarly, a temporal pattern could not be identified that indicated a consistent trend of increasing or decreasing fish IBI value over the nine-year monitoring period at any of the three sites. However, on a temporal basis the fish IBI values during the nine-year monitoring period indicate the fish community within the reconstructed reach to be equivalent to the undisturbed upstream location. For example, the fish IBI value at site GLC in the reconstructed reach was higher in May 1993, April 1997 (Table 1), and August of 1997 (Table 2) than calculated for undisturbed upstream site GLA during previous fish survey events. The fish IBI results show the fish community within the reconstructed reach of Galum Creek has supported a fish community of equal health and integrity as the undisturbed upstream reach since 1988. Furthermore, the fish IBI values for the downstream reach were similar and suggest any stream reconstruction and relocation activities had negligible effect on downstream fish communities.

The findings of a comparison of the fish IBI results for Galum Creek to the fish IBI status of the regional reference site ATE-06 in the Eagle Creek Basin indicate slightly higher IBI scores for Galum Creek during both April and August. Based on IBI scores the fish community within reconstructed reach of Galum Creek is comparable to a fish community typical of a relatively undisturbed stream within Illinois. Confounding factors associated with the recalculation of the fish IBI data and evaluation of the fish IBI results includes uncertainty if a comparable level of effort and methods were used during sampling efforts from both streams, and the relative flow conditions at the time of each survey event.

3.2.2 Bonnie Creek Fish IBI

The Bonnie Creek 1997 fish IBI values for April (Table 1) were slightly higher than IBI values determined for August (Table 2) at each of the upstream, reconstructed reach, and downstream monitoring sites. The decrease in IBI value from spring to late summer was minimal and within a range of values that indicated no meaningful change in health and integrity of the fish community. In April, the IBI value for the upstream site BCA was 32 compared to a value of 30 for site BCC in the reconstructed reach; and in August the IBI values were 26 and 28 for sites BCA and BCC, respectively. The downstream site BCD has the lowest IBI value of 27 for the April survey (Table 1) compared to the same value of 28 as the reconstructed reach during August (Table 2). No spatial pattern among the sites was consistent for both the April and August sample events. In addition, the difference in IBI value among the sites was not of sufficient magnitude to indicate a meaningful difference in fish community health among the sites during either April or August. The 1997 fish IBI data indicates that the fish community health and integrity in the reconstructed reach of Bonnie Creek was equivalent to the fish community health and integrity found upstream at the undisturbed BCA site.

The Bonnie Creek fish IBI values for 1997 ranged from 26 to 32 and were higher at all three monitoring sites than the fish IBI value of 24 calculated for the fish survey data from ATE-06 in the Eagle River Basin. Based on the conclusion by Matson and Hite (1987) that site ATE-06 is generally not impacted by regional mining activities and local land use, the low fish IBI results for ATE-06 may indicate typical results for a small Illinois stream. The comparison to Bonnie Creek fish IBI results would also indicate the reconstructed reach of Bonnie Creek supports a fish community typical of a relatively undisturbed stream within Illinois. Confounding factors associated with the evaluation of the fish IBI data includes uncertainty whether a comparable level of effort and method (shock time, double pass, block nets) was used during all sampling efforts, flow conditions at the time of sampling, and the time span of eleven years between survey events from ATE-06 and Bonnie Creek.

3.3 Benthic Macroinvertebrates

Benthic macroinvertebrate samples were collected coincident with fish survey events at Galum Creek, Bonnie Creek, and White Walnut Creek. Macroinvertebrate samples from ATE-06 in the Eagle Creek Basin were collected on October 20 and 22, 1986. Thus, a comparison to the August sampling from the reconstructed stream reaches is most appropriate considering the developmental life stages and size of benthic organisms susceptible for capture. Flow conditions in Upper Eagle Creek were described as dry with the exception of one extensive pool area at the time of sample collection (Matson and Hite 1987), which likely mimicked the zero to near zero flow conditions in the reconstructed streams in August and September when benthos samples were collected. Sample collection methods were assumed to be equivalent among all sampled streams and incorporated a D-net kick sample and subsequent picking of organisms from sampled rocks, net, and debris (Galum, Bonnie, and White Walnut Creek's) or use of a standard 30-mesh screen or D-net with subsequent picking of organisms at ATE-06. Following collection, samples were preserved and transported to an analytical laboratory for species identification and enumeration. Replicate samples were not discussed and did not appear to be collected from any site.

The combined assemblage of benthic organisms reported from the reconstructed stream samples represented various snails, aquatic isopods and amphipods, flat worms, mussels and clams, as well as the major groups of aquatic insects common to most of the central United States. Sample total abundance was variable and for the late summer collections in the reconstructed streams often resulted in samples with less than 100 specimens; below the typical guideline of a minimum 100 specimens and a target of 300 specimens for determining a benthic index of biotic integrity. With the exception of the August 1988 sample from GLD in Galum Creek (dominated by chironomid flies) and two other samples of 124 and 159 organisms, all other samples contained less than 100 specimens. The late summer benthic collections (August or September) contained a higher abundance of organisms than the spring (April or May) collections, an expected pattern that can be attributed to life stage development and larger body sizes for the benthic community in general. Similarly, total taxonomic richness (number of different taxa in the sample) was generally higher in the late summer collections than reported for the spring samples. Macroinvertebrate richness was variable from season to season and from year to year depending upon the stream. Macroinvertebrate richness in Bonnie Creek and Galum Creek tended to decreased downstream in both the summer and fall collections prior to 2003.

Specific to the objective of this evaluation is the status and response of the biological communities in the reconstructed reaches of Bonnie Creek, Galum Creek and White Walnut Creek. Evaluation of spatial and temporal trends is a useful approach for identifying biological patterns and responses. Spatial trends for the reconstructed portions of the streams can be identified by the repeated occurrence of patterns observed for comparisons of the biological data from undisturbed sites with the reconstructed reach. The undisturbed upstream sample site would be the primary target for comparison since the undisturbed downstream sample site may have been influenced by stream reconstruction activities. Temporal trends for the reconstructed portions of the stream can be identified by a evaluation of year-to-year changes in biological patterns within the reconstructed reach, which can also be compared to similar year-to-year changes for the undisturbed upstream sample site.

Spatial and temporal pattern recognition for the entire period of monitoring in Bonnie Creek and Galum Creek were limited by presence/absence data reported for monitoring results in 2003 and 2006. Macroinvertebrate community metrics that provided the most information with respect to implications for community health and structure that incorporated presence/absence data included total taxonomic richness and EPT (Ephemeroptera, Plecoptera and Trichoptera) richness. Total taxonomic richness has implications to diversity and stability of the community and EPT richness has implications to water quality conditions as these organisms as a group are generally considered sensitive to a variety of pollutants. Higher EPT richness and total richness values are typically interpreted as an indicator of more favorable stream conditions that can include better water quality, a more diverse physical habitat and stable hydrologic patterns. EPT richness and total richness values for Bonnie Creek, Galum Creek, and White Walnut Creek sample events are presented in Table 3.

The temporal biological response to reconstruction of the stream channel will focus on identifying trends or patterns in the macroinvertebrate IBI (MIBI) when possible, and patterns for EPT richness and total richness that increase and are indicative of favorable stream conditions. The implications of increasing MIBI, EPT richness, and total richness values over time can suggest improved water quality conditions, stability of the physical characteristics of the channel, and development of a balanced biotic community. Progression towards stable physical characteristics and the development of a balanced biotic community is a natural process in newly formed streams.

3.3.1 Galum Creek Macroinvertebrate Richness

Galum Creek macroinvertebrate data indicated EPT richness to be low at all sites surveyed that suggested a complex benthic community was likely not present. In general, there was some seasonal variation in EPT richness at all sites with the lowest EPT richness values often observed during the August sample periods (Table 3). For example, the average EPT richness at the undisturbed upstream GLA site for the 1988, 1993, 1997, 2003 and 2006 sampling periods was 4 species during April compared to an average of 2 species in August. EPT richness was often lower within the reconstructed reach that observed at the undisturbed upstream site, but this pattern was not consistent. For example, average EPT richness in the reconstructed stream reach of Galum Creek for the same sampling period was lower (1 species in April) but equivalent to the upstream GLA site in August with an average of 2 species. Considering the low values for EPT richness in Galum Creek (Table 3), these differences between seasons and sites are negligible and can be accounted for by sample collection and sample analysis variability.

Total species richness was also considered low and variable; exhibited an inconsistent seasonal trend for lower total richness values for the August samples; and similar to EPT richness generally indicated a complex benthic community was not present. During the April monitoring period total richness averaged 12 species at the undisturbed upstream site compared to an average of 8 species in the reconstructed reach of Galum Creek, but showed little difference in average total richness for the August samples (Table 3). Average total richness for the August samples was 14 species for both the upstream GLA site and the reconstructed reach of Galum Creek. The maximum total richness value at the upstream GLA site for any given sample period was 19 species and comparable to the maximum of 18 species for the reconstructed reach of Galum Creek. Similar to the EPT richness, the observed spatial differences in total richness values were not of sufficient magnitude to be considered meaningful and likely could be accounted for by variability in sample collection and sample analysis.

Galum Creek data includes five sampling periods spanning 18 years. The strongest temporal pattern includes an overall increase in total richness for samples collected in April in the reconstructed reach (GLC) and the undisturbed upstream GLA site. Total richness during April increased from 3 to 17 species in the reconstructed reach of Galum Creek over the 1988 to 2006 monitoring period, but the increase in richness primarily occurred after 2003 (Table 3). The downstream total richness data indicated that until 2006, richness in the relocated portion of Galum Creek was suppressed during April compared the rest of the stream system. In contrast, during August the EPT richness and total richness values from the relocated reach were similar to the undisturbed upstream and downstream sites over the entire 18-year monitoring period (Table 3). Based on macroinvertebrate richness data alone samples from the relocated reach of Galum Creek were equivalent with samples from the undisturbed upstream site and downstream site as early as August 1998.

The available Galum Creek data includes a 9-year time span from 1988 to 1997 when species abundance values were recorded prior to presence/absence data reporting in 2003 and 2006. Subjecting the 1988, 1993, and 1997 macroinvertebrate abundance data to the IBI protocols of Plafkin et al. (1988) calculation of the MIBI incorporates additional community metrics other than richness-based metrics and allows another evaluation of trends that includes functional aspects of the assemblages. Higher MIBI values indicate better healthy biotic communities and imply higher quality habitat and water conditions and MIBI results for Galum Creek are presented below in Section 3.4.

3.3.2 Bonnie Creek Macroinvertebrates

In general the Bonnie Creek EPT richness and total richness values were low and variable, and indicated a complex macroinvertebrate community was likely not present in Bonnie Creek. A spatial comparison of total richness and EPT richness values from the reconstructed reach with the upstream undisturbed site (BCA) indicated a general pattern of lower richness values in the reconstructed reach during April. This was most apparent for the EPT richness metric, which also exhibited lower richness values for the August monitoring period (Table 3). For example, the average EPT richness at BCA during the April monitoring period for 1997, 2003 and 2006 was 5 species compared to an average EPT richness of 2 species for the August samples from BCA. For the same time periods average EPT richness from the reconstructed reach was 4 species in April and 2 species for the August samples. The maximum EPT richness for the April samples was 5 species at both the upstream site and reconstructed reach in April 2006.

Bonnie Creek total richness values for the reconstructed reach compared to the undisturbed upstream site showed a pattern similar to EPT richness, but less apparent. Average total richness for 1997, 2003 and 2006 at BCA was 13 species for both the April and August sampling periods, compared to mean total richness values of 11 species (April) and 12 species (August) from the reconstructed stream reach. The variability in total richness and EPT richness that occurred from sampling period to sampling period at the upstream site was typically mirrored at the reconstruction stream reach (Table 3). Differences in the EPT richness and total richness values among sites and sample periods were considered negligible relative to the overall level of species richness reported for the Bonnie Creek samples.

Temporal patterns in the reconstructed portion of Bonnie Creek were also not well established. The strongest pattern was best demonstrated by a relatively stable increase in total richness value from 12, to 11, to 16 species exhibited by the August samples collected in 1997, 2003, and 2006 (Table 3). However, there is an overall pattern of reduced richness in the macroinvertebrate communities for the 2003 samples that curtail the presence of a progressive trend. This same pattern for richness values in 2003 is present in samples collected from Galum Creek suggesting a widespread meteorological or hydrologic event may have occurred. The minor increases of only 1 or 2 species in value for EPT richness and total richness between 1997 and 2006 in the reconstructed portion of Bonnie Creek may be attributed to sampling and analysis variability and are of insufficient magnitude to be meaningful. Based on macroinvertebrate richness data the monitoring results indicate the macroinvertebrate community from the relocated reach of Bonnie Creek was equivalent with the macroinvertebrate community from an undisturbed upstream site.

3.3.3 White Walnut Creek Macroinvertebrates

Evaluation of spatial patterns in White Walnut Creek was based on the April and August sampling periods in 2003. EPT richness was higher in April than observed in August at all sites, and found to be equivalent between the reconstructed reach and the undisturbed upstream site (Table 3). EPT richness at both the upstream and reconstructed reach sites was 5 species in April 2003. Total richness was also higher in April than observed in August and observed to be higher in the relocated reach of Bonnie Creek than for the undisturbed site upstream during both sampling periods. The higher EPT richness and total richness values in the reconstructed portion of White Walnut Creek indicates benthic community conditions equivalent or better than occur upstream, but may also reflect successful colonization by a large suite of organisms within new available habitats. Intense colonization commonly occurs in new stream habitats and elevated richness values are frequently observed during initial monitoring events. Elevated richness values indicating intense colonization were not as apparent during the initial monitoring events of 1997 in Bonnie Creek or during 1988 in Galum Creek (Table 3).

Temporal trends in White Walnut Creek cannot be ascertained because only one year of presence/absence macroinvertebrate monitoring data is available.

3.4 Macroinvertebrate IBI

The macroinvertebrate IBI (MIBI) based on Plafkin et al. (1988) included benthic organism abundance data from site ATE-06 collected in October 1986 in the Eagle Creek Basin (Matson and Hite 1987) as a reference location and for comparison to organism abundance data from Bonnie Creek (1997) and Galum Creek (1988, 1993, and 1997) for samples collected in April or May (spring) and August or September (late summer). MIBI results for Bonnie Creek in 2003 and 2006; Galum Creek for 2003 and 2006, and White Walnut Creek for 2003 cannot be determined because of the presence/absence type of data presented. Results will focus on a comparison of the MIBI score from site ATE-06 to MIBI scores for the reconstructed reaches of Bonnie Creek and Galum Creek. An IBI score of less that 83% of the reference condition is interpreted as indication of less biological integrity (Plafkin et al. 1988). Macroinvertebrate IBI (MIBI) metric values, metric scores, and MIBI value for site ATE-06, and sites in Bonnie Creek and Galum Creek are presented for April sample collections in Table 4 and for August sample collections in Table 5.

3.4.1 Galum Creek MIBI

MIBI results for samples collected from the reconstructed reach in Galum Creek during spring and late summer of 1988, 1993, and 1997 showed seasonal differences when compared to the reference site macroinvertebrate assemblage. During spring, the MIBI values from the reconstructed reach of Galum Creek were 18, 36, and 26 for 1988, 1992, and 1997, respectively, (Table 4) indicating that only in spring of 1993 the macroinvertebrate assemblage was equivalent with the reference site ATE-06 conditions (85.7%). The MIBI results also indicated the upstream undisturbed site supported macroinvertebrate communities were equivalent to the reference condition only in spring of 1997 (also 85.7%). However, because of the expected seasonal difference in life stage of aquatic insects within streams the comparison of the April Galum Creek samples to the October ATE-06 samples may not be entirely reliable. The MIBI results for the late summer macroinvertebrate collections are likely a more appropriate and reliable comparison to the October ATE-06 reference samples. MIBI results for the late summer samples for 1988, 1992, and 1997 (Table 5) from the reconstructed reach in Galum Creek were 36, 38, and 40, or 85.7% and considered equivalent with the reference at ATE-06. The August results include conditions in 1988 when the MIBI value for the undisturbed upstream site in Galum Creek was not equivalent to the reference condition (66.7% of ATE-06).

The results of the August MIBI for Galum Creek indicates the relocated reach supported a macroinvertebrate community equivalent with the undisturbed upstream reach and equivalent with a regional reference site considered minimally impacted by mining activities (Table 4). Furthermore, the results of the MIBI evaluation suggest the recovery of the macroinvertebrate community in Galum Creek occurred within a 5-year time span and continued to improve. Confounding factors with respect to conclusions regarding the reference site include seasonal variation as demonstrated by comparisons to the regional reference site using the April Galum Creek macroinvertebrate collections.

3.4.2 Bonnie Creek MIBI

MIBI results for samples collected from the undisturbed upstream portion of Bonnie Creek in 1997 were 94% (Table 4) and 85% (Table 5) of the MIBI results of the regional reference MIBI score of 42 at the ATE-06 site. This comparison demonstrated Bonnie Creek was equivalent with a typical small stream in Illinois. Within the reconstructed portion of Bonnie Creek, the MIBI value was 34 for both April (Table 4) and August (Table 5) sample events. The MIBI data for the reconstructed portion of Bonnie Creek for both April and August 1997 monitoring periods was 81% of the regional reference site ATE-06, but nearly identical to the undisturbed upstream reach in Bonnie Creek. These data indicate the benthic community from the relocated reach in Bonnie Creek recovered to a level that was equivalent with undisturbed reaches upstream of the reconstruction zone. It is possible that the normal condition of the Bonnie Creek benthic community exhibits an MIBI value more typical of agriculture influences and related land use practices or hydrologic patterns.

4 Conclusions

The fish and benthic macroinvertebrate communities from reconstructed and relocated portions of Bonnie Creek, Galum Creek, and White Walnut Creek (Perry County, Illinois) were compared to benthic and fish samples from upstream undisturbed sites from each stream, and a selected non-impacted regional Illinois stream site from Eagle Creek Basin surveyed by IEPA (Matson and Hite 1988). All study streams are first or second order small streams from geographical regions with current agricultural land uses and previously supported coal mining. The non-impacted regional reference stream selected for comparison is conservative as localized agricultural land use and impacts are minimal. The purpose of the comparison was to determine the status of the biological communities in the reconstructed and relocated portions of each stream and assess whether the reconstruction stream reaches supported benthic and fish communities that were equivalent to normal stream or regional reference conditions.

The fish community evaluation was based on the fish IBI following Simon and Dufour (1998) using abundance based data from the sites. The macroinvertebrate community evaluation included comparison of the EPT richness and total richness metrics for spatial and temporal patterns because some of the available data was not abundance data and presented as species presence/absence data. In addition, the macroinvertebrate Index of Biotic Integrity (MIBI) following Plafkin et al. (1989) was used for available abundance data to assess overall status among sites.

The fish assemblage at all streams and sites evaluated exhibited species common to small streams of Illinois. The streams were characterized by a dominance of green, bluegill, and longear sunfish, along with frequent capture of red shiner and sand shiner (Bonnie Creek) and/or blackstripe and bluntnose minnow (Galum Creek and White Walnut Creek). A fish IBI evaluation could not be conducted for White Walnut Creek because only species presence/absence data were available. Conclusions based on the result of this evaluation of the fish community survey data include the following:

- Fish IBI values for all streams evaluated, including the reference stream, were in the low range of possible values and indicated communities dominated by tolerant species that were primarily omnivores and capable of exploiting a variety of physical habitats.
- Fish IBI values for Galum Creek indicate the reconstructed reach to support a fish community typical of a relatively undisturbed stream within Illinois, and difference in fish IBI value among the undisturbed upstream reach and the reconstructed site was not of sufficient magnitude to indicate a meaningful spatial difference in fish community health or integrity.
- Fish IBI values for Bonnie Creek in 1997 indicated fish community health and integrity in the reconstructed reach was equivalent to the fish community health and integrity of the undisturbed upstream site BCA. The comparison to Bonnie Creek fish IBI results would also indicate the reconstructed reach of Bonnie Creek supports a fish community typical of a relatively undisturbed stream within Illinois.

The benthic macroinvertebrate assemblage at all sites included species common to small streams in Illinois and represented most of the major groups of aquatic insects as well as snails, aquatic isopods and amphipods, flat worms, mussels and clams. Conclusions based on the evaluation of richness based comparisons using the EPT richness and total richness metrics, and comparison using MIBI values include the following:

- Overall abundance (number of specimens) of benthic macroinvertebrate organisms in the samples collected, including the regional reference location may be an artifact of differences in sampling effort. Confounding factors at the time of sample collection included lack of measureable flow at many sites. However, low specimen counts were common to all sites and collection dates in this study and was not considered a significant factor in interpretation of results.
- The richness-based metrics for all stream sites were highly variable. Comparisons indicated a general, but not conclusive, trend for higher EPT richness in April samples compared to samples collected in August. In contrast, the total richness metric showed a general, but not conclusive, trend for higher richness in August than present in April. Both the EPT richness and total richness values were low and any differences in richness value were not consistent or of sufficient magnitude to be meaningful and likely could be attributed to sample collection and sample analysis variability, and the influence of the agricultural setting for the sample streams.
- The results of the August MIBI for Galum Creek indicated the reconstructed reach attained macroinvertebrate community recovery that reflects normal in-stream conditions (as depicted by MIBI values for the undisturbed upstream site). MIBI results also indicated the Galum Creek macroinvertebrate assemblage was equivalent to a reference site considered minimally impacted by mining activities. Results of the MIBI evaluation suggest the recovery of the macroinvertebrate community in Galum Creek occurred within a 5-year time span.
- MIBI results for samples collected from the reconstructed portion of Bonnie Creek in 1997 indicated recovery to a level equivalent with MIBI values from the undisturbed upstream reach of Bonnie Creek. Total richness and EPT richness values at the upstream, relocated reach, and downstream monitoring sites were relatively consistent for the 1997, 2003, and 2006 monitoring periods in Bonnie Creek and suggested the normal condition of the benthic macroinvertebrate community in Bonnie Creek may be typical of streams in agricultural settings and slightly lower than determined for the un-impacted regional reference site selected.

5 References

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Tables

TABLE 1. ILLINOIS STREAM MONITORING SITE SUMMARY TABLE

Stream	Site	Lat.	Long.	County
Galum	GLA	38.0854833	-89.555033	Perry
	GLC	38.0575833	-89.5559	Perry
	GLD	38.0473833	-89.521	Perry
Bonnie	BCA	38.0865667	-89.514617	Perry
	BCB2	38.0821667	-89.514617	Perry
	BCB3	38.0648667	-89.520017	Perry
	BCB4	38.0559333	-89.524867	Perry
	BG	38.0494333	-89.5226	Perry
White Walnut	WWA	38.121	-89.306	Perry
	WWC2	38.113	-89.326	Perry
	WWD	38.109	-89.348	Perry
Eagle Creek Basin	ATE-06	37.62889	-89.326	Saline

TABLE 2. APRIL-MAY FISH INDEX OF BIOLOGICAL INTEGRITY METRIC VALUES AND METRIC SCORES FOR RECONSTRUCTED STREAMS IN PERRY COUNTY, ILLINOIS AND A REFERENCE SITE IN THE EAGLE CREEK BASIN

IBI Metric	Eagle Creek	Bonnie Creek			Galum Creek			Galum Creek			Galum Creek		
	ATE-06	BCA	BCC	BCD	GLA	GLC	GLD	GLA	GLC	GLD	GLA	GLC	GLD
	20-Oct-86		Apr-97			Apr-98			May-93			Apr-97	
Metric Values													
Total Number of Species[1]	7	8	9	4	7	6	12	14	8	12	7	5	8
Number of Darter Species[2]	0	0	0	0	0	0	0	0	0	0	0	0	0
Number of Sunfish Species[3]	4	3	2	2	4	2	3	4	4	3	3	3	3
Number of Sucker Species[3]	0	0	0	0	0	1	0	1	0	0	0	0	0
Number of Sensitive Species[4]	1	3	1	1	1	2	4	3	1	3	2	1	2
% Tolerant Individuals[3]	95	46.6	38.2	43.5	24.4	2.0	40.0	45.5	37.9	41.8	17.2	42.2	26.9
% Omnivores[3,8]	37	14.7	2.2	0	0	0	3.3	28.4	0	10.1	12.1	1.6	15.3
% Insectivores[3,8]	53	85.3	97.4	100	92.7	98.0	94.4	67.0	75.9	86.1	87.9	98.4	84.7
% Carnivores[3,8]	11	0	0	0	7.3	2.0	2.2	3.4	24.1	38.0	0	0	26.3
Catch per Unit Effort[5]	unknown												
% Simple Lithophils[3,8]	0	0	1.8	0	0	0	3.3	0	0	1.3	0	0	1.9
% DELT Anomolies[5,8]	0												
Number of Minnow Species[6]	2	4	6	2	0	0	3	4	0	5	3	1	5
% Pioneer Species[7,8]	37.0	16.4	7.0	32.3	19.5	0	33.3	34.1	24.1	38.0	17.2	42.2	26.3
% Darter, Madtom, Sculpin[2]	0	0	0	0	0	0	0	0	0	0	0	0	0
Metric Scores													
Total Number of Species[1]	3	3	3	1	1	1	3	5	3	3	1	1	3
Number of Darter Species[2]	1	1	1	1	1	1	1	1	1	1	1	1	1
Number of Sunfish Species[3]	5	3	3	2	5	3	3	5	5	3	3	3	3
Number of Sucker Species[3]	1	1	1	1	1	1	1	1	1	1	1	1	1
Number of Sensitive Species[4]	1	3	1	1	1	1	3	3	1	3	1	1	1
% Tolerant Individuals[3]	1	3	3	3	5	5	3	3	3	3	5	3	3
% Omnivores[3,8]	1	5	5	5	1	5	5	3	1	5	5	5	5
% Insectivores[3,8]	1	5	5	5	1	5	5	5	1	5	5	5	5
% Carnivores[3,8]	1	1	1	1	3	1	1	1	5	1	1	1	1
Catch per Unit Effort[5]	3	3	3	3	3	3	3	3	3	3	3	3	3
% Simple Lithophils[3,8]	1	1	1	1	1	1	1	1	1	1	1	1	1
% DELT Anomolies[5,8]	5	3	3	3	1	3	3	3	1	3	3	3	3
Fish IBI score	24	32	30	27	24	30	32	34	26	32	30	28	30
Flow (cfs)		9.8	6.3	15.9	*	<1	1	13	25	31	9	16	18
Number of Minnow Species[6]	1	3	3	1	1	1	3	3	1	3	1	1	3
% Pioneer Species[7,8]	1	5	5	3	1	5	3	3	1	3	5	3	3
% Darter, Madtom, Sculpin[2]	1	1	1	1	1	1	1	1	1	1	1	1	1

1 Figure 5 in Simon and Dufour 1998
2 Figure 6 in Simon and Dufour 1998
3 Table 3 in Simon and Dufour 1998
4 Figure 11 in Simon and Dufour 1998
5 Unknown. Assume middle value for catch per unit effort, and no deformities for all sites.
6 Figure 9 in Simon and Dufour 1998
7 Table 2 in Simon and Dufour 1998
8 May be adjusted for low number of fish captured

TABLE 3. AUGUST FISH INDEX OF BIOLOGICAL INTEGRITY METRIC VALUES AND METRIC SCORES FOR RECONSTRUCTED STREAMS IN PERRY COUNTY, ILLINOIS AND A REFERENCE SITE IN THE EAGLE CREEK BASIN

	Eagle Creek	Bonnie Creek			Galum Creek			Galum Creek			Galum Creek		
IBI Metric	ATE-06	BCA	BCC	BCD	GLA	GLC	GLD	GLA	GLC	GLD	GLA	GLC	GLD
	20-Oct-86	Aug-97			Aug-88			Aug-93			Aug-97		
Metric Values													
Total Number of Species[1]	7	12	6	4	12	7	10	10	9	7	9	9	13
Number of Darter Species[2]	0	0	0	0	0	0	0	0	0	0	1	0	0
Number of Sunfish Species[3]	4	3	1	3	4	2	3	4	2	2	3	3	3
Number of Sucker Species[3]	0	0	0	0	2	0	0	1	0	0	0	0	0
Number of Sensitive Species[4]	1	2	1	1	1	2	2	2	1	1	2	2	3
% Tolerant Individuals[3]	95	63.6	20.9	88.3	13.7	4.7	7.7	34.1	50	44.9	19.6	48.6	32.7
% Omnivores[3,8]	37	29.0	0.0	0.0	3.9	4.7	1.5	9.8	26.3	5.1	0	0	12.3
% Insectivores[3,8]	53	68.5	100	95.0	92.2	81.4	93.8	87.8	68.4	94.4	92.8	89.2	75.9
% Carnivores[3,8]	11	1.9	0.0	5.0	2	14	4.6	0.0	5.3	0	7.2	10.8	11.7
Catch per Unit Effort[5]	unknown												
% Simple Lithophils[3,8]	0	3.1	6.0	0	5.9	0	0	2.4	0	1	1	0	11.7
% DELT Anomolies[5,8]	0												
Number of Minnow Species[6]	2	6	4	0	2	2	2	2	1	4	1	1	5
% Pioneer Species[7,8]	37	49.4	0	88.3	9.8	0	3.1	24.4	18.4	40.8	17.5	35.1	31.5
% Darter, Madtom, Sculpin[2]	0	0	0	0	0	0	0	0	0	0	5.2	0	0
Metric Scores													
Total Number of Species[1]	3	3	1	1	3	1	3	3	3	3	3	3	5
Number of Darter Species[2]	1	1	1	1	1	1	1	1	1	1	1	1	1
Number of Sunfish Species[3]	5	3	1	3	5	3	3	5	3	3	3	3	3
Number of Sucker Species[3]	1	1	1	1	3	1	1	1	1	1	1	1	1
Number of Sensitive Species[4]	1	1	1	1	1	1	1	1	1	1	1	1	3
% Tolerant Individuals[3]	1	1	5	1	5	5	5	3	3	3	5	3	3
% Omnivores[3,8]	1	3	5	5	5	1	5	1	1	5	5	1	5
% Insectivores[3,8]	1	5	5	5	5	1	5	1	1	5	5	1	5
% Carnivores[3,8]	1	1	1	3	1	5	1	1	3	1	3	5	5
Catch per Unit Effort[5]	3	3	3	3	3	3	3	3	3	3	3	3	3
% Simple Lithophils[3,8]	1	1	1	1	1	1	1	1	1	1	1	1	1
% DELT Anomolies[5,8]	5	3	3	3	3	1	3	1	1	3	3	1	3
Fish IBI score	24	26	28	28	36	24	32	22	22	30	34	24	38
Flow (cfs)		0	0	2.8	**	**	<1	*	1	2	**	***	***
Number of Minnow Species[6]	1	3	3	1	1	1	1	1	1	3	1	1	3
% Pioneer Species[7,8]	1	3	5	1	5	1	5	1	1	3	5	1	3
% Darter, Madtom, Sculpin[2]	1	1	1	1	1	1	1	1	1	1	1	1	1

1 Figure 5 in Simon and Dufour 1998
2 Figure 6 in Simon and Dufour 1998
3 Table 3 in Simon and Dufour 1998
4 Figure 11 in Simon and Dufour 1998
5 Unknown. Assume middle value for catch per unit effort, and no deformities for all sites.
6 Figure 9 in Simon and Dufour 1998
7 Table 2 in Simon and Dufour 1998
8 May be adjusted for low number of fish captured

TABLE 4. BENTHIC MACROINVERTEBRATE RICHNESS VALUES SUMMARY FOR RECONSTRUCTED STREAMS IN PERRY COUNTY, ILLINOIS.

Site	Bonnie Creek					
	Spring			Fall		
	Apr-97	Apr-03	Apr-06	Aug-97	Aug-03	Aug-06
EPT Richness						
BCA	9	3	5	2	2	2
BCC	6	1[a]	5[a]	2	2[a]	3[a]
BCD	7	0	1	na	1	1
Total Richness						
BCA	15	10	16	15	14	11
BCC	13	6[a]	16[a]	12	11[a]	14[a]
BCD	13	7	13	na	12	10

Site	White Walnut Creek	
	Apr-03	Aug-03
EPT Richness		
WWA	5	0
WWC	5	2
WWD	3	3
Total Richness		
WWA	13	7
WWC	15	10
WWD	13	16

Site	Galum Creek									
	Spring					Fall				
	Apr-88	Apr-93	Apr-97	Apr-03	Apr-06	Aug-88	Aug-93	Aug-97	Aug-03	Aug-06
EPT Richness										
GLA	1	5	6	1	5	1	2	5	3	3
GLC	1	2	2	1[a]	3[a]	3	2	1	2[a]	1[a]
GLD	4	2	5	0	4	4	5	1	2	2
Total Richness										
GLA	7	14	13	9	17	15	13	19	11	14
GLC	3	8	7	7[a]	17[a]	15	14	14	14[a]	18[a]
GLD	10	7	11	8	19	19	12	12	14	27

1. "a" indicates number of different taxa from multiple monitoring sites sampled in 2003 and 2006 in the reconstructed reach.

TABLE 5. APRIL-MAY BENTHIC MACROINVERTEBRATE INDEX OF BIOLOGICAL INTEGRITY (MIBI) METRIC VALUES AND METRIC SCORES FOR RECONSTRUCTED STREAMS IN PERRY COUNTY, ILLINOIS AND A REFERENCE SITE IN THE EAGLE CREEK BASIN

MIBI Metric	Eagle Creek	Bonnie Creek			Galum Creek			Galum Creek			Galum Creek		
	ATE-06	BCA	BCC	BCD	GLA	GLC	GLD	GLA	GLC	GLD	GLA	GLC	GLD
	20-Oct-86	Apr-97			Apr-88			May-93			Apr-97		
Flow (cfs)		9.8	6.3	15.9	NA	1.0	1.0	12.7	24.9	0.85	8.9	15.7	18.1
Conductivity (umhos/cm)		523	694	3050	1100	2200	4100	477	1289	1305	562	1510	1470
Metric Values													
Taxa richness	10	15	13	13	7	3	10	14	8	7	13	7	11
Hilsenhoff Biotic Index	7.0	4.6	5.7	5.4	5.9	5.5	5.7	6.2	6.7	4.9	6.0	5.8	5.3
Scrapers:filt/collectors	0.4	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2
EPT: chironomid	1.6	5.4	4.7	0.8	0.0	1.0	1.1	0.0	3.3	34.0	1.7	0.0	2.5
% Dominant Taxa	27.2	26.8	46.3	48.0	37.5	50.0	40.9	41.0	24.1	58.1	30.2	41.9	27.7
EPT richness	2	9	6	7	1	1	4	5	2	2	6	2	5
Community Loss	reference	0.53	0.69	0.76	1.50	3.00	0.80	0.43	1.10	1.20	0.62	1.10	0.81
Shredders:Total	0.00	0.04	0.01	0.07	0.00	0.00	0.18	0.05	0.00	0.67	0.00	0.00	0.13
Metric Scores													
Taxa richness	6	6	6	6	4	0	6	6	4	4	6	4	6
Hilsenhoff Biotic Index	6	6	6	6	6	6	6	6	6	6	6	6	6
Scrapers:filt/collectors	6	2	0	0	0	0	0	0	0	0	0	0	2
EPT: chironomid	6	6	6	4	0	4	4	0	6	6	6	0	6
% Dominant Taxa	6	4	0	0	2	0	0	0	4	0	2	0	4
EPT richness	6	6	6	6	0	0	6	6	6	6	6	6	6
Community Loss	6	4	4	4	4	2	4	6	4	4	4	4	4
Shredders:Total	0	6	6	6	6	6	6	6	6	6	6	6	6
MIBI SCORE	42	40	34	32	22	18	32	30	36	32	36	26	40
Percent of Reference		95.2	81.0 *	76.2	52.4	42.9 *	76.2	71.4	85.7	76.2	85.7	61.9 *	95.2

* Score <83% and indicates reconstructed reach has lower biotic integrity than reference condition.

TABLE 6. AUGUST BENTHIC MACROINVERTEBRATE INDEX OF BIOLOGICAL INTEGRITY (MIBI) METRIC VALUES AND METRIC SCORES FOR RECONSTRUCTED STREAMS IN PERRY COUNTY, ILLINOIS AND A REFERENCE SITE IN THE EAGLE CREEK BASIN.

MIBI Metric	Eagle Creek ATE-06 20-Oct-86	Bonnie Creek Aug-97			Galum Creek Aug-88			Galum Creek Aug-93			Galum Creek Aug-97		
		BCA	BCC	BCD	GLA	GLC	GLD	GLA	GLC	GLD	GLA	GLC	GLD
Flow (cfs)		<1.0	<1.0	2.0	<1	<1.0	1.0	<1.0	0.85	2.0	<1.0	<1.0	<1.0
Conductivity (umhos/cm)		620	1350	4780	395	3150	4050	597	2080	2190	720	2480	2630
Metric Values													
Taxa richness	10	15	12	NA	15	15	19	13	14	11	19	14	12
Hilsenhoff Biotic Index	7.0	7.3	6.1	NA	7.5	6.8	6.3	6.6	6.5	5.5	6.0	5.9	5.2
Scrapers:filt/collectors	0.4	0.0	0.0	NA	0.0	0.0	0.1	0.0	0.0	0.1	4.0	2.0	0.0
EPT: chironomid	1.6	2.1	3.1	NA	0.9	5.0	0.4	3.0	12.0	3.1	0.5	1.0	27.8
% Dominant Taxa	27.2	41.7	43.5	NA	38.8	43.9	52.0	18.4	26.8	32.3	33.3	24.2	69.8
EPT richness	2	2	2	NA	1	3	5	2	2	5	5	1	1
Community Loss	reference	0.47	0.58	NA	0.53	0.47	0.37	0.62	0.57	0.81	0.42	0.64	0.66
Shredders:Total	0.00	0.00	0.00	NA	0.01	0.00	0.00	0.00	0.00	0.15	0.04	0.00	0.00
Metric Scores													
Taxa richness	6	6	6	NA	6	6	6	6	6	6	6	6	6
Hilsenhoff Biotic Index	6	6	6	NA	6	6	6	6	6	6	6	6	6
Scrapers:filt/collectors	6	0	0	NA	0	0	0	0	0	2	6	6	0
EPT: chironomid	6	6	6	NA	4	6	2	6	6	6	2	2	6
% Dominant Taxa	6	0	0	NA	2	0	0	6	4	2	2	2	0
EPT richness	6	6	6	NA	0	6	6	6	6	6	6	6	0
Community Loss	6	6	4	NA	4	6	6	4	4	4	6	6	4
Shredders:Total	0	6	6	NA	6	6	6	6	6	6	6	6	6
MIBI SCORE	42	36	34	NA	28	36	32	40	38	38	40	40	28
Percent of Reference		85.7	81.0 *	NA	66.7	85.7	76.2	95.2	90.5	90.5	95.2	95.2	66.7

* Score <83% and indicates reconstructed reach has lower biotic integrity than reference condition.

Figures

FIGURE 1. View of Regional Reference Site ATE-06 in the Eagle Creek Basin, Saline County, Illinois.

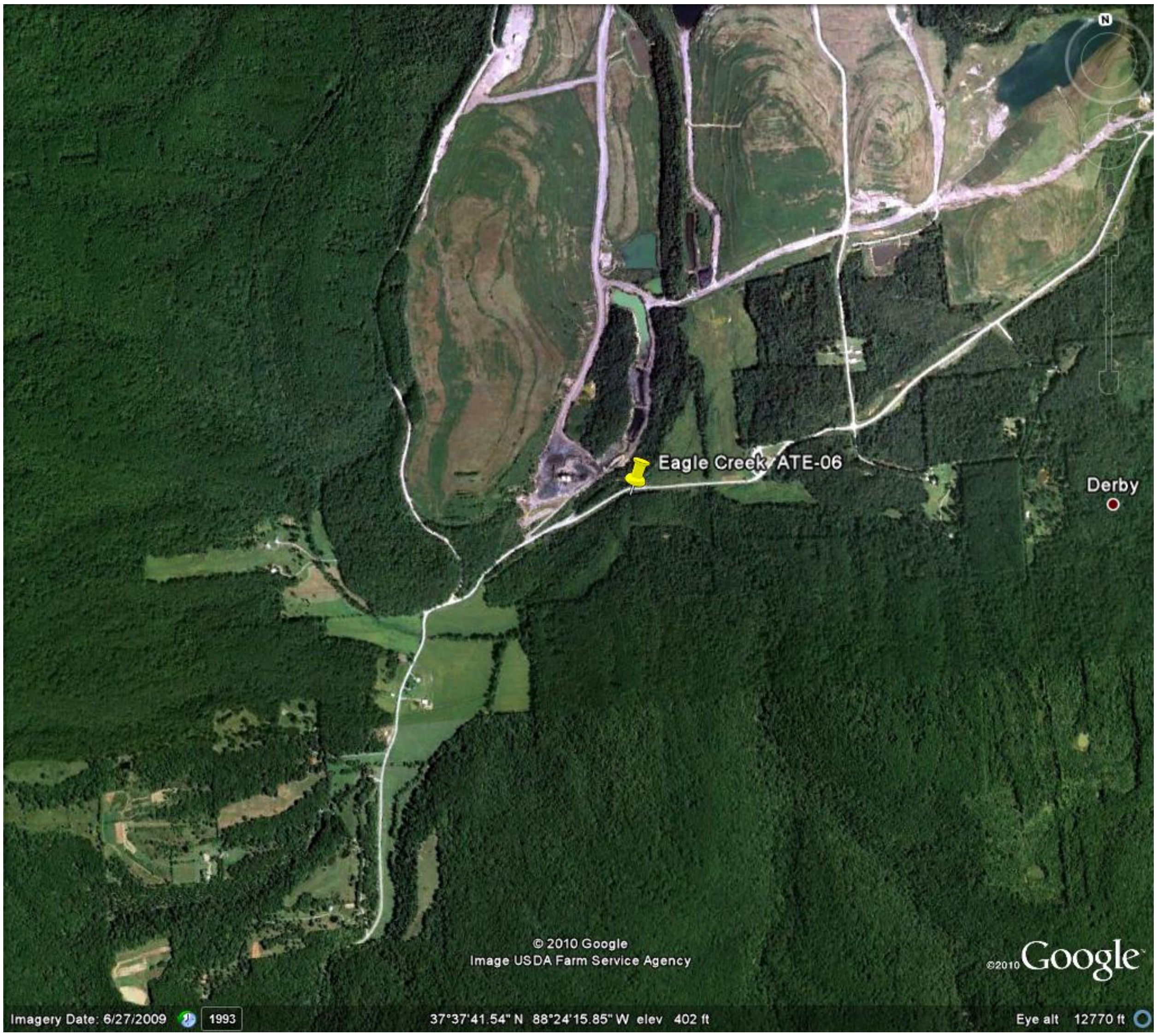

FIGURE 2. View of Galum and Bonnie Creek Monitoring Sites, Perry County, Illinois.

FIGURE 3. View of White Walnut Creek Monitoring Sites, Perry County, Illinois.

APPENDIX B – EXHIBIT 2

Impacts of Coalmine Discharges on Illinois Unionid Mussels

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Introduction

While active coalmines are subject to National Pollutant Discharge Elimination System (NPDES) permits requiring periodic monitoring of the chemical content and/or toxicity of discharges, little is known about the extent to which these monitoring techniques are protective of indigenous freshwater mussels (Unionidae). As an example, Belanger et al. (1990) noted that only one laboratory study of acute copper toxicity or chronic impairment to freshwater bivalves was included in the United States Environmental Protection Agency's (U.S. EPA, 1985) ambient water quality criteria document for Cu, and those data were not used in criteria calculation. Cherry et al. (2002) recently conducted a study to determine if effluent permit limits for copper at a coal-fired power plant located on the Clinch River, Virginia, were protective of native unionids. They tested the toxicity of Cu to 17 genera, including 15 that were indigenous to the Clinch River, as well as the standard U.S. EPA effluent testing organisms, *Ceriodaphnia dubia* (a cladoceran) and *Pimephales promelas* (the fathead minnow). Seven of the 10 most sensitive genera to copper in this study were freshwater mussels, including the top three. *C. dubia* ranked sixth, after four mussels and a mayfly, and the fathead minnow ranked fourteenth. This study provided evidence that mussels may be among the most sensitive aquatic organisms to contaminants, and that standard bioassessment techniques (e.g., water column toxicity testing with *C. dubia*) and WQC standards may not be protective of them.

The objective of this study was to conduct various bioassessment techniques to determine if the discharge from the Riola Mine was causing measurable impairment to instream communities, particularly to mussels.

Methods

<u>Water/Sediment Chemistry</u>

Water chemistry analysis included general wet chemistry (temperature, pH, dissolved oxygen, conductivity, alkalinity and hardness) and metals analysis. Temperature, pH, dissolved oxygen, and conductivity were measured in the field, while alkalinity and hardness (mg/L as $CaCO_3$) were determined in the laboratory by titration (APHA, 1998). Water samples were analyzed for total concentrations of Al, Ca, Cd, Cu, Fe, magnesium (Mg), manganese (Mn), Na, lead (Pb), and zinc (Zn) by Inductively-Coupled Plasma (ICP) spectrometry at the Illinois State Water Survey, Champaign, IL.

Laboratory Water Column Toxicity Testing

Water column samples will be collected from each sampling station to test for toxicity to the cladoceran, *Ceriodaphnia dubia*. *C. dubia* has been cultured house according to U.S. EPA methods (1993) in the Soucek laboratory at the Illinois Natural History Survey since 2002. Prior to testing, organisms were fed a diet of *Selenastrum capricornutum* and a Yeast-Cereal Leaves-Trout Chow (YCT) mixture at a rate of 0.18 ml each per 30-ml water, daily. Cultures were maintained at 25±1ºC, and a 16:8 (L:D) photoperiod.

Range finding tests were conducted for each site (upstream and downstream of discharge) to determine if acute or chronic toxicity testing was appropriate. For range finding tests, five organisms were placed into each of four replicate 50-ml beakers containing undiluted water collected from each station for 48 h. Moderately hard, reconstituted (EPA^{100}) water was used as the control. At the end of the test period, percent survival for each station was recorded. Results of range finding tests indicated that further testing (i.e., generation of LC50s) was not required.

Laboratory Sediment Toxicity Testing

Whole-sediment samples were collected from each site according to standard methods (U.S. EPA, 2001) using a polyethylene scoop. Whole sediments were tested for toxicity to the midge, *Chironomus tentans*, according to U.S. EPA methods (1994

Benthic Macroinvertebrate Community Sampling

Benthic macroinvertebrate surveys were conducted concurrent with the mussel transplant test according to the U.S. EPA Rapid Bioassessment Protocols (RBP) with slight modifications (Barbour et al., 1999). Modifications of RBP protocols included the fact that riffle, run, pool and shoreline rooted areas were sampled for 5 minutes per site using dip-nets with 800-µm mesh dipnets rather than for 20 “jabs” with a 500-µm mesh dipnet. One composite sample was collected per site according to RBP recommendations. All organisms in each sample were identified to the lowest practical taxonomic level (usually genus) using standard keys (Merritt and Cummins, 1996; Pennak, 1989). Chironomids were identified as either subfamily Tanypodinae or non-Tanypodinae.

Caged Transplanted Mussel Study

To experimentally evaluate the toxicity of mining discharges to freshwater mussels, fatmuckets (*Lampsilis siliquoidea*) were collected from a nearby reference site within the same drainage (Little Vermillion River at CR 600 E, north of Sidell) for transplant studies. Organisms were collected, transported to the laboratory, sexed, measured for shell length (anterior to posterior margin) and depth (dorsal to ventral margin) and wet mass. Mussels then were secured in plastic cages, which were subdivided and marked to allow identification of individuals before and after deployment. Approximately 2 days after collection of mussels, cages were transported back to the field sites and secured to the substrate using re-bar. Five cages were placed at each monitoring station in Fayette Drain (upstream and downstream of the Riola mine property) and each cage contained one male and one female mussel. Cages were monitored on a weekly basis to prevent filling with sediments. After 60 days, cages were removed from the sampling stations, organisms were transported back to the laboratory, and shell length, wet mass, tissue condition index (dry tissue mass divided by dry shell mass, Newton et al., 2001). Dried soft tissues were digested for metals analysis according to U.S. EPA (1991) methods, and tissue digestates were submitted to the Illinois State Water Survey for analysis using ICP spectrometry. Tissues from both mussels in a given cage were combined for digestion and analysis, resulting in five mussel tissue samples per sampling station.

RESULTS

Riola Mine Discharge, Fayette Drain, near Georgetown, IL.

According to IDNR staff, the Black Beauty Riola Coal underground coalmine, located in Vermilion County, with both open and reclaimed gob/caked slurry coal waste facilities was opened in 1994 on the Fayette Drain. Fayette Drain is a tributary to the Little Vermilion River, which is known to support populations of the State-listed endangered Little Spectaclecase, *Villosa lienosa*, and the State-threatened Slippershell, *Alasmidonta viridis*. An IDNR survey crew in August 2001 found 18 *V. lienosa* and 2 *A. viridis*, along with individuals of four other species, including the formerly State-listed Pondhorn, *Uniomerus tetralasmus*, less than one mile downstream of the Riola Mine outfall within a channelized portion of the stream. Listed species comprised 80% of live mussels found. The Riola Mine received a modified NPDES permit in late 2001.

During a site visit on September 2, 2002, a sample of the discharge was collected and analyzed for metals and general water chemistry. General water chemistry was as follows: pH 7.5, conductivity 1,450 µmhos/cm, alkalinity 100 mg/L (as $CaCO_3$), hardness 230 mg/L (as $CaCO_3$). This discharge sample was analyzed for total metal concentrations by inductively coupled plasma spectrophotometry at the Illinois State Water Survey. These concentrations are shown in Table 1. Eight priority toxic pollutants with Water Quality Criteria (WQC) for protection of aquatic life (U.S. EPA, 1999) were measured (beryllium, cadmium, chromium, copper, lead, nickel, selenium, and zinc) and with the exception of zinc, all were below detection limits. Zinc was substantially below its WQC limit of 0.240 mg/L (at hardness of 230 mg/L as $CaCO_3$). Five non-priority pollutant metals (U.S. EPA, 1999) were analyzed (aluminum, barium, boron, iron, and manganese), all with concentrations below respective WQC for protection of aquatic life or human health. **The discharge was not flowing during other site visits.**

Table 1. Total metal concentrations in water sample collected from the Black Beauty Riola Mine discharge into the Fayette Drain, Vermillion County, IL. The sample was collected directly from the discharge pipe, not from the stream. BDL = below detection limit. All concentrations in mg/L (ppm) total metal.

metal	concentration	metal	concentration
Al	0.063	Mo	0.009
B	0.296	Na	378
Ba	0.083	Ni	bdl
Be	bdl	P	bdl
Ca	61.8	Pb	bdl
Cd	bdl	S	92.1
Co	bdl	Sb	0.029
Cr	bdl	Se	bdl
Cu	bdl	Si	0.869
Fe	0.024	Sn	bdl
K	6.65	Sr	0.183
Li	0.019	V	0.019
Mg	18.0	Zn	0.001
Mn	0.005		

Laboratory Water Column and Sediment Toxicity Testing

Water samples collected from upstream and downstream reaches, and from the discharge itself caused no acute toxicity to *Ceriodaphnia dubia* on any occasion. In fact no test organisms died at all as a result of exposure to collected water column samples. Likewise with whole sediment toxicity tests with *Chironomus tentans*, samples collected from upstream and downstream sites did not cause significant mortality or growth reduction to the midge larvae.

Benthic Macroinvertebrate Community Sampling

A total of 2,562 individuals belonging to 36 different taxa were collected at both sites (Table 2). At the upstream site, 24 taxa were identified, while 36 were identified at the downstream site. The upstream site was dominated by dipterans (mostly of the family Chironomidae) and hemipterans (Corixidae), whereas the downstream site was dominated by both gastropod and bivalved (Pelecypoda) mollusks (Fig. 1). Two different genera of bivalves were found at the downstream site: *Pisidium* (252 specimens) and *Sphaerium* (22 specimens), both fingernail clams. Only four individuals of the genus *Pisidium* were found at the upstream site. Each site had the same three genera of mayflies (*Caenis* sp., *Hexagenia* sp., and *Baetis* sp.), but 113 mayflies were collected at the downstream site whereas only 31 were collected upstream of the discharge.

Table 2. Complete list of organisms collected using RBP methods both upstream and downstream of the Riola Mine discharge site. Yellow shading indicates a taxon not present at one site but collected at the other.

			upstream	downstream
Family	Taxon	common name	# of specimens	# of specimens
Annelids				
	Hirudinea	leech	1	0
	Oligochaeta	segmented worm	78	28
Mollusks				
Pelycypoda/Veneroida				
Sphaeriidae	*Pisidium*	fingernail clam	4	252
Sphaeriidae	*Sphaerium*	fingernail clam	0	22
Gastropoda				
Lymnaeidae	*Fossaria*	snail	1	100
Lymnaeidae	*Pseudosuccinea*	snail	0	1
Physidae	*Physella*	snail	133	334
Planorbidae	Planorbidae	snail	0	3
Crustaceans				
Malacostraca/Decapoda				
Cambaridae	*Orconectes propinquus*	crayfish	16	21
Cambaridae	*Procambarus acutus*	crayfish	4	16
Malacostraca/Amphipoda				
Hyalellidae	*Hyalella azteca*	scud (amphipod)	1	2
Malacostraca/Isopoda				
Asellidae	Caecidotea	aquatic sowbug (isopod)	91	9
Insects				
Ephemeroptera				
Caenidae	*Caenis*	square-gilled mayfly	19	86
Ephemeridae	*Hexagenia*	burrowing mayfly	11	1
Baetidae	*Baetis*	swimming mayfly	1	26
Odonata				
Calopterygidae	*Calopteryx*	damselfly	6	5
Coenagrionidae	*Enallagma*	damselfly	8	16
Gomphidae	*Dromogomphus*	dragonfly	1	7
Libellulidae	*Plathemis lydia*	dragonfly	0	11
Hemiptera				
Corixidae	juveniles	water boatman	235	38
Gerridae		water strider	0	1
Trichoptera				
Leptoceridae	*Oecetis*	caddisfly	0	2
Lepidoptera				
Pyralidae	*Acentria*	aquatic moth	0	1
Coleoptera				
Haliplidae	*Peltodytes*	crawling water beetle	13	70
Elmidae	*Dubiraphia*	riffle beetle	56	26

Gyrinidae	*Gyrinus*	whirligig beetle	0	3
Hydrophilidae	*Tropisternus*	water scavenger beetle	1	3
Dytiscidae	*Agabus*	predaceous diving beetle	2	17
Dytiscidae	*Laccophilus*	predaceous diving beetle	0	2
Dytiscidae	*Rhantus*	predaceous diving beetle	2	0
Dytiscidae	*Derrovatellus*	predaceous diving beetle	0	4
Diptera				
Tabanidae	*Chrysops*	deer fly	1	7
Ceratopogonidae	*Bezzia*	No-see-um or biting midge	15	3
Chironomidae		non-biting midge	657	81
Simuliidae		black fly	0	2
Stratiomyidae		soldier fly	0	1
Sciomyzidae		marsh fly	0	4
	total abundance		1357	1205
	total richness		24	35
	EPT richness		3	4

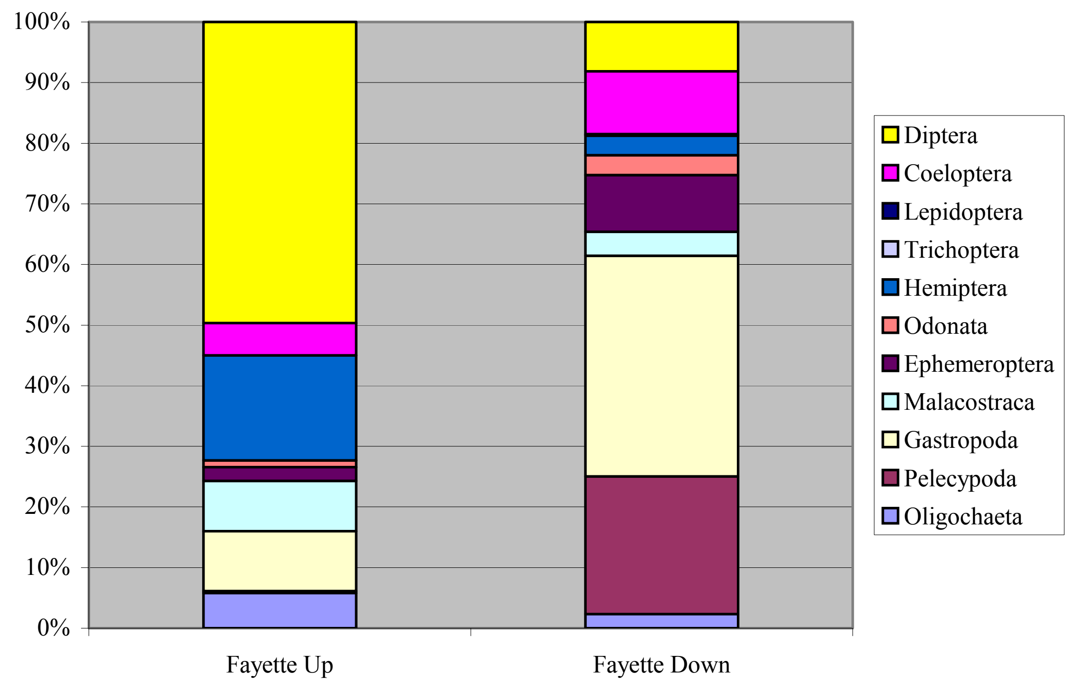

Figure 1. Taxonomic composition of benthic communities at sampling sites upstream and downstream of the Riola Mine discharge point in Fayette Drain. Bars show percent abundance of various taxa, e.g. the bright yellow portion of each bar indicates the percentage of the total number of organisms collected that belong to the insect Order Diptera.

Caged Transplanted Mussel Study

At the end of 60 days, mussels caged and transplanted to sites upstream and downstream of the Riola mine discharge were evaluated for fitness using various body measurements (Table 3). Mussels were similar in whole body weight, length (anterior to posterior margin), depth (dorsal to ventral margin), dry tissue weight and shell weight when comparing means of all mussels at a given site. That is, the average values of these parameters for upstream mussels were not significantly different (p>0.05) from those for mussels transplanted downstream of the discharge. However, despite the fact that organisms collected from the Little Vermillion River were selected from a specific size range, variability in these parameters before transplantation may reduce the ability to statistically detect differences due to the discharge. Therefore, an additional normalizing parameter called Tissue Condition Index (TCI) was calculated. This index is a measure of the relative fitness of the organism because it divides the mass of the dried soft parts of the organism by the mass of the dry shell, i.e., it is a measure of the robustness of the organisms. This normalized parameter is more appropriate for comparing fitness of organisms upstream and downstream of the discharge. As shown in table 2, the mean TCI value of the mussels at the downstream site was nominally lower than that for the upstream mussels (higher TCI suggests better condition); however, these means were not significantly different (p = 0.2373). Therefore, it appears that site of transplantation had no effect on fitness of the caged mussels.

Table 3. Various body measurements of mussels (*Lampsilis siliquoidea*) after being transplanted upstream and downstream of the Riola mine discharge in Fayette Drain for 60 days. Individual measurements and averages are shown.

Upstream						
Individual	Weight (mm)	Length (mm)	Depth (mm)	Dry Tissue Weight	Shell Weight	TCI
1	90.38	80.52	48.68	2.06	51.18	4.02
2	94.59	84.85	48.92	2.15	46.47	4.62
3	110.02	83.90	53.01	2.72	57.09	4.77
4	99.87	90.52	53.38	2.65	48.69	5.44
5	96.01	83.78	52.11	2.76	51.54	5.36
6	89.43	84.41	51.39	2.46	43.80	5.61
7	102.45	85.20	51.39	2.84	50.65	5.62
8	96.90	86.94	51.32	3.04	48.76	6.24
9	80.49	77.91	49.49	1.83	41.81	4.37
10	77.31	83.57	49.02	2.60	38.35	6.77
average	93.74	84.16	50.87	2.51	47.83	5.28
std dev	9.83	3.38	1.73	0.39	5.42	0.85
Downstream						
Individual	Weight (mm)	Length (mm)	Depth (mm)	Dry Tissue Weight	Shell Weight	TCI

1	124.74	89.81	53.20	2.63	62.35	4.21
2	113.88	88.58	50.81	2.73	61.94	4.40
3	96.69	83.48	54.25	1.70	49.04	3.47
4	96.87	86.87	50.53	2.64	49.29	5.36
5	76.23	79.53	48.11	2.05	39.01	5.24
6	79.29	86.75	48.31	2.32	39.23	5.92
7	92.58	78.29	51.03	2.24	48.99	4.57
8	92.60	90.66	53.98	2.55	45.77	5.56
9	dead	n/a	n/a	n/a	n/a	n/a
10	94.98	85.27	51.14	2.29	48.70	4.70
average	96.43	85.47	51.26	2.35	49.37	4.83
std dev	15.16	4.33	2.22	0.33	8.29	0.76

Generally, elemental concentrations in dried mussel tissues were similar for organisms transplanted upstream and downstream of the discharge. Table 4 shows average concentrations at each site. Concentrations were significantly different ($p < 0.05$) for only four elements. For three of these (Al, Se, and Ti) the mean concentration was significantly lower downstream of the discharge than upstream. The only element that was significantly higher downstream of the discharge was Ba, and the toxicology of this element is relatively unclear. One finding of interest was that tissue Cu concentrations at both stations were near or above 8 ppm (mg/kg), which is a level thought to cause sublethal effects, such as loss of copper regulation, in zebra mussels, *Dreissena polymorpha* (U.S. EPA, 2000). However, the upstream value (9 ppm) was higher than the downstream value (7.5 ppm) so these concentrations should not be attributed to the discharge.

Table 4. Elemental concentrations in tissues (mg/kg dry weight) of fatmuckets (*Lampsilis siliquoidea*, Unionidae) transplanted in the Fayette Drain upstream and downstream of the Riola Mine discharge near Georgetown, IL. Yellow shading indicates means are significantly different ($p < 0.05$).

		upstream		downstream
element		average (n = 5)		average (n = 4)
Al	mg/kg	551.6		382.2
B	mg/kg	343.3		337.8
Ba	mg/kg	464.9		537.9
Ca	mg/kg	64461.5		71738.3
Cd	mg/kg	2.7		2.7
Co	mg/kg	3.1		2.7
Cr	mg/kg	3.9		4.7
Cu	mg/kg	9.0		7.5

Fe	mg/kg	2316.0		2502.0
K	mg/kg	2637.8		2480.4
Li	mg/kg	2.1		2.0
Mg	mg/kg	2299.3		2368.5
Mn	mg/kg	5097.7		5561.5
Na	mg/kg	1718.4		1847.5
Ni	mg/kg	10.8		3.9
P	mg/kg	34506.8		38225.0
Pb	mg/kg	8.4		8.8
S	mg/kg	6120.5		6204.4
Se	mg/kg	15.0		11.4
Si	mg/kg	1448.4		1346.0
Sr	mg/kg	88.4		97.9
Ti	mg/kg	11.6		8.1
V	mg/kg	2.8		2.8
Zn	mg/kg	440.4		489.4

Conclusions—Riola Mine Discharge

Based on the data generated in this study, including water chemistry, toxicity testing, benthic macroinvertebrate sampling, and bioaccumulation studies with transplanted freshwater mussels, it appears that the discharge from the Black Beauty Riola is not having an adverse impact on the biota inhabiting the Fayette Drain. Benthic macroinvertebrate communities were more diverse downstream of the mine site and were dominated by mollusks, whereas the upstream site was dominated by rather tolerant midges (Chironomidae). None of the water or sediment samples collected were toxic to standard tests organisms. Furthermore, transplanted mussels did not accumulate significant levels of harmful metals in their tissues compared to upstream transplants after two months exposure, and their TCI values were not significantly lower than those placed upstream of the mine site. Water column and sediment toxicity testing may represent snapshots, in that if they indicate toxicity or lack thereof, it is only for the moment at which samples were collected; however, benthic macroinvertebrate surveys and mussel transplants are not. The latter two techniques, particularly benthic macroinvertebrate sampling, represent longer-term assessments of the condition of a particular site. In this case, all types of data collected were in agreement in suggesting that the discharge is not a major factor in structuring the aquatic communities of Fayette Drain.

The remaining locations did not have active discharges at the time of the study and therefore the data generated are representative of background conditions at the sites.

White County--Pattiki Coal Mine Waste Disposal Facility/Little Wabash River

A coal mine waste disposal facility near Carmi is proposed for an unnamed tributary of the Little Wabash River several miles above its confluence with the Wabash River. There is no other past or present coal mining in the watershed. At this point, the Little Wabash River is known to support populations of the State-listed threatened spike, *Elliptio dilatata*, and the federally-listed fat pocketbook, *Potamilus capax*.

The discharge tributary was located and study sites set up upstream and downstream of it. An excellent source population of mussels was located in the Little Wabash River upstream of the city of Carmi near First Street. The dominant species found in this community is *Quadrula quadrula*, the mapleleaf, and specimens were collected and caged in late August. Cages were deployed at the study sites on August 27 and retrieved on October 9, 2002. To our knowledge, the tributary receiving the discharge was dry during the mussel exposure period. However, data from this first experiment will provide insight into background metal levels and mussel condition values under low flow. I have received no information regarding whether or not the NPDES permit has been approved.

Laboratory Water Column and Sediment Toxicity Testing
Water samples collected from upstream and downstream reaches, and from the discharge itself caused no acute toxicity to *Ceriodaphnia dubia*. Likewise with whole sediment toxicity tests with *Chironomus tentans*, samples collected from upstream and downstream sites did not cause significant mortality or growth reduction to the midge larvae.

Benthic Macroinvertebrate Community Sampling
A total of 130 individuals belonging to 13 different taxa were collected at both sites (Table 5). At the upstream site, 8 taxa were identified, while 10 were identified at the downstream site. The upstream site was dominated by mayflies (Ephemeroptera) and dipterans (mostly of the family Chironomidae), whereas the downstream site was dominated by both gastropods (Pleuroceridae) and mayflies. The upstream site had four genera of mayflies, whereas only two genera were collected at the downstream site; however, more individuals from the order Ephemeroptera were collected at the downstream site.

Table 5. Complete list of organisms collected using RBP methods both upstream and downstream of the Pattiki Coal Mine Waste Disposal Facility discharge stream in the Little Wabash River near Carmi, IL. Yellow shading indicates a taxon not present at one site but collected at the other.

		Upstream	Downstream
Family	Taxon	# of specimens	# of specimens
Annelids			

	Hirudinea	0	1
	Oligochaeta	0	2
Mollusks			
Gastropoda			
Pleuroceridae		0	38
Crustaceans			
Malacostraca/Decapoda			
Cambaridae	*Orconectes sp.*	1	0
Palaemonidae	*Palaemonetes kadiakensis*	2	3
Malacostraca/Isopoda			
Asellidae	Caecidotea	1	1
Insects			
Ephemeroptera			
Caenidae	*Caenis*	3	2
Ephemeridae	*Hexagenia*	1	0
Heptageniidae	*Stenonema*	11	34
Tricorythidae	*Tricorythodes*	1	0
Odonata			
Coenagrionidae	*Argia*	0	4
Coleoptera			
Elmidae	*Stenelmis*	0	3
Diptera			
Chironomidae		20	2
	total abundance	40	90
	total richness	8	10
	EPT richness	4	2

Caged Transplanted Mussel Study

Mussels were caged within 200 meters upstream and downstream of the tributary designated to receive the discharge. In addition, at the end of the caging experiment, 10 specimens of the same species were collected from the initial collection site (upstream of Carmi near First Street) to make the same measurements. This was done to determine if caging of mussels has an impact on their condition. As shown in table 6, there was no difference in final wet weight, length (measured as the distance from the anterior to the posterior end), or depth (measured as the distance from the dorsal to the ventral margin) at the end of the experiment between the caged mussels placed upstream and downstream of the discharge tributary. However, the mussels that were caged and transplanted had lower average weights, lengths, and depths than the "native" or un-caged mussels collected at the end of the experiment. In spite of this finding, there was no difference among any of the groups in TCI, indicating that their overall condition was similar, despite differences in size. These results indicate that placement upstream or downstream of the discharge tributary had no impact on overall organism health. Furthermore, caging and transplanting did not have a negative impact on organism health. This is encouraging, because it indicates that when there is an environmental stressor present, its effects will not be masked by effects of taking mussels out of their natural environment and placing them in cages.

Table 6. Size and condition measurements for caged and uncaged mapleleafs, *Quadrula quadrula*, upstream and downstream of the Pattiki Coal Mine Waste Disposal Facility discharge stream in the Little Wabash River near Carmi, IL. Different capital letters indicate significant differences among means ($p < 0.05$).

Site	Wet Weight (g)	Length (mm)	Depth (mm)	TCI*
Upstream (n = 15)	210.8 ± 22.7 B	88.0 ± 3.5 B	72.9 ± 2.8 B	3.1 ± 0.3 A
Downstream (n = 14)	188.4 ± 21.9 B	85.2 ± 2.2 B	69.8 ± 2.2 B	3.3 ± 0.5 A
Native (n = 10)	257.3 ± 31.3 A	93.9 ± 2.1 A	76.4 ± 2.7 A	3.0 ± 0.5 A

* TCI= tissue condition index, measured as dry weight of soft tissue divided by dry weight of shell, multiplied by 100.

Generally, elemental concentrations in dried mussel tissues were similar for organisms transplanted upstream and downstream of the discharge. Table 7 shows average concentrations at each site. Concentrations were significantly different ($p < 0.05$) for only two elements, Cd and Mg. For both Cd and Mg, the mean concentration was highest in the naitve (uncaged) organisms. The reason for the significant decrease in these metal concentrations in caged mussels is unknown. None of the other elemental concentrations were outstanding.

Table 7. Elemental concentrations in tissues (mg/kg dry weight) of mapleleafs (*Quadrula quadrula*, Unionidae) transplanted in the Little Wabash River upstream and downstream of the Pattiki Coal Mine Waste Disposal Facility discharge stream near Carmi, IL. Yellow shading indicates means are significantly different ($p < 0.05$).

		upstream	downstream	native
element	units	average (n = 5)	average (n = 5)	average (n = 3)
Ag	mg/kg	bdl	bdl	bdl
Al	mg/kg	32.4	33.2	33.6
As	mg/kg	2	1.9	2.1
Ba	mg/kg	472	487	500
Ca	mg/kg	29,200	31,600	31,333
Cd	mg/kg	0.4	0.5	0.8
Co	mg/kg	0.9	0.9	1.0
Cr	mg/kg	1.4	1.2	1.4

Cu	mg/kg	6.9	6.1	6.7
Fe	mg/kg	1,840	1,660	1,900
Mg	mg/kg	944	984	1,066
Mn	mg/kg	7560	8020	8,133
Na	mg/kg	940	1,029	1,193
Ni	mg/kg	7.2	3.9	4.8
P	mg/kg	24,600	25,000	26,333
Pb	mg/kg	0.3	0.3	0.3
Se	mg/kg	2	2	2
Sr	mg/kg	91.1	100.2	100.4
Zn	mg/kg	101.1	85.8	98.2

Clinton County--Monterey Coal Co. #2 Mine/Kaskaskia River

This underground coal mine located near Albers is closed, but has a coal waste facility that pumps treated ground water to avoid contamination of an aquifer, which is the principal water supply for surrounding communities. The mine has recently applied for an NPDES permit which would allow it to discharge treated water to the Kaskaskia River, just below its confluence with Shoal Creek, where it is designated as an INAI Site because it is a high-diversity (10+ species) mussel stream. No listed species are known to occur in this vicinity. No other coal mining is known to have occurred in this immediate area.

According to Mr. Jack Rickner of ExxonMobil at the Monterey #2 mine, the discharge site had not been definitively determined at the time of the study, but proposed locations include into Shoal Creek just above its confluence with the Kaskaskia River or into the Kaskaskia itself, just below Shoal Creek. I contacted the owner of the land adjacent to these sites and was granted access to the property where Shoal Creek enters the Kaskaskia River. We located a sufficient source population of mussels in the Kaskaskia drainage for use in these experiments. The site was ~4 miles northwest of Tuscola, IL, at county road 1450. The most abundant species at this site is *Amblema plicata*, the threeridge, and we collected sufficient numbers for the experiment. Cages were deployed on September 20, 2002, and removed on November 6, 2002. Mussels were also caged at the collection site near Tuscola for the same duration.

Laboratory Water Column and Sediment Toxicity Testing
Water samples collected from upstream and downstream reaches, and from the discharge itself caused no acute toxicity to *Ceriodaphnia dubia*. As with the previously described sites, whole sediment samples collected from upstream and downstream sites did not cause significant mortality or growth reduction to the midge larvae, *Chironomus tentans*.

Benthic Macroinvertebrate Community Sampling
A total of 211 individuals belonging to 8 different taxa were collected at both sites (Table 8). At the upstream site, 6 taxa were identified, 6 were identified at the downstream site as well. Both sites were dominated by hemipterans in the family Corixidae, but baetid

mayflies were present at both sites. Both sites had extremely poor habitat for most sensitive benthic macroinvertebrates as the channels were deeply cut, and the bottom substrate consisted of either shifting sands or sediment composed largely of clays.

Table 8. Complete list of organisms collected using RBP methods in Shoal Creek and the Kaskaskia River near Albers, IL.

		Shoal Creek	Kaskaskia @ Albers
Family	Taxon	# of specimens	# of specimens
Crustaceans			
Malacostraca/Decapoda			
Palaemonidae	*Palaemonetes kadiakensis*	0	2
Malacostraca/Amphipoda			
Hyalellidae	*Hyalella azteca*	1	0
Insects			
Ephemeroptera			
Baetidae	*Baetis*	19	4
Odonata			
Coenagrionidae	*Argia*	0	4
Hemiptera			
Corixidae	*Trichocorixa*	100	47
Coleoptera			
Elmidae	*Ancyronyx*	1	0
Dytiscidae	*Derrovatellus*	1	0
Diptera			
Chironomidae		25	7
	total abundance	147	64
	total richness	6	6
	EPT richness	1	1

<u>Caged Transplanted Mussel Study</u>

Because this discharge is still in the "pending" stage, the experiment was conducted at these sites to obtain background levels for comparison when the discharge is implemented. Five cages containing three mussels each were placed in the Kasakaskia River immediately downstream of Shoal Creek, near Albers, IL, and in the mouth of Shoal Creek. Both of these sites are potential locations for the Monterey discharge. In addition, organisms were caged and placed back in the Kaskaskia River at the site at which they were collected (near Tuscola, IL). This was done to compare the responses of caged mussels at their collection site to those of organisms transplanted a large distance from the collection point. As shown in table 9, all of the caged animals had similar weights, lengths and depths at the end of the experimental period; however, significant differences ($p < 0.05$) were observed in average TCI at the end of the experiment. Specifically, organisms placed at the original collection site and in Shoal Creek, had similar average TCI, but these averages were significantly less than the average TCI of the organisms placed in the Kaskaskia River at Albers. Because shell measurements were similar at all three sites, this difference is attributed to greater soft tissue mass in the organisms placed in the Kaskaskia at Albers. The river is much larger, in terms of discharge, at this point than it is at the collection site near Tuscola, IL, and is also much larger than Shoal Creek. It is possible that the larger discharge site is more enriched with fine particulate organic matter (FPOM) and dissolved organic carbon (Vannote et al., 1980), thus, providing a greater source of filterable food material for the caged mussels. The fact that the mussels transplanted in Shoal Creek were in similar condition to those placed back at their collection site indicates that water quality in Shoal Creek is sufficient at this time to support mussel populations. However, the physical habitat quality in Shoal Creek and in the Kaskaskia River downstream of Shoal Creek is less than optimal for freshwater unionids.

Table 9. Size and condition measurements for caged and uncaged threeridges, *Amblema plicata*, in the Kaskaskia River drainage near Albers, and Tuscola, IL. Different capital letters indicate significant differences among means ($p < 0.05$).

Site	Weight (g)	Length (mm)	Depth (mm)	TCI*
Kaskaskia, Albers (n = 14)	358.6 ± 57.2 A	119.9 ± 4.5 A	83.0 ± 3.7 A	2.8 ± 0.9 A
Shoal Creek (n = 15)	387.6 ± 64.9 A	120.9 ± 7.7 A	83.6 ± 3.8 A	2.4 ± 0.4 B
Kaskaskia, Tuscola (n = 14)	363.7 ± 46.8 A	119.4 ± 5.9 A	82.6 ± 4.8 A	2.1 ± 0.3 B

* TCI= tissue condition index, measured as dry weight of soft tissue divided by dry weight of shell, multiplied by 100.

Of the 19 elements measured in the mussel tissues from these sites, 15 were highest in the mussels caged at their original collection location near Tuscola, and in the case of 10 of these, concentrations were significantly higher ($p < 0.05$) at the Tuscola location than in the Kaskaskia River near Albers location (Table 10). Metals in tissues of mussels caged in Shoal Creek were generally lower than those in the Kaskaskia Tuscola location, but often not significantly lower. It appears that being caged at the Clinton County locations for two months allowed the mussels to depurate some of the metals contained in their tissues before transplantation.

Table 10. Elemental concentrations in tissues (mg/kg dry weight) of threeridges (*Amblema plicata*, Unionidae) transplanted in the *Amblema plicata*, in the Kaskaskia River drainage near Albers, and Tuscola, IL. Yellow shading indicates means are significantly different ($p < 0.05$) from cells not shaded or shaded blue. Cells shaded green are not significantly different from either yellow or blues cells.

		Shoal Ck	Kaskaskia Albers	Kaskaskia Tuscola
element		average (n = 5)	average (n = 5)	average (n = 5)
Ag	mg/kg	2	3.5	4.1
Al	mg/kg	332	138	211
As	mg/kg	3.7	5.6	7.2

Ba	mg/kg	1220	998	1380
Ca	mg/kg	76,400	65,200	87,400
Cd	mg/kg	0.7	0.5	0.6
Co	mg/kg	1.4	1.4	1.8
Cr	mg/kg	3.8	3.6	5
Cu	mg/kg	4.6	4.5	4.3
Fe	mg/kg	2,820	2,321	2,900
Mg	mg/kg	3320	2778	3,620
Mn	mg/kg	8420	6897	8,760
Na	mg/kg	1560	1,300	1,400
Ni	mg/kg	6.1	5.3	6.2
P	mg/kg	53,200	45,503	56,400
Pb	mg/kg	2.5	2.3	2.8
Se	mg/kg	3.3	2.5	3.7
Sr	mg/kg	302	254	368
Zn	mg/kg	490	432	560

Logan County--Sandra Miller Bellrose Nature Preserve/Sugar Creek

This privately owned site is designated an INAI Site (Sugar Creek-Salt Creek) high-mussel diversity stream (at least 17 species present) and has received approval as an Illinois Nature Preserve. The site is known to contain remarkably dense mussel populations in its 0.8-mile segment, but, interestingly, no State-listed species are present.

Laboratory Water Column and Sediment Toxicity Testing

Water samples collected from upstream and downstream reaches, and from the discharge itself caused no acute toxicity to *Ceriodaphnia dubia*. As with the previously described sites, whole sediment samples collected from upstream and downstream sites did not cause significant mortality or growth reduction to the midge larvae, *Chironomus tentans*.

Benthic Macroinvertebrate Community Sampling

A total of 629 individuals belonging to 24 different taxa were collected at Sugar Creek (Table 11). This site also had the highest EPT richness of any site sampled in the study, with five genera of mayflies and three genera of caddisflies. Five genera of non-unionid mollusks were also collected at this site.

Table 11. Complete list of organisms collected using RBP methods in Sugar Creek, Sandra Miller Bellrose Nature Preserve drainage, Logan Co., IL.

		Sugar Creek
Family	Taxon	# of specimens

Annelids		
	Oligochaeta	5
Mollusks		
Pelycypoda/Veneroida		
Sphaeriidae	*Pisidium*	3
Sphaeriidae	*Sphaerium*	3
Corbiculidae	*Corbicula*	45
Gastropoda		
Pleuroceridae	*Pleurocera*	11
Lymnaeidae	*Fossaria*	1
Crustaceans		
Malacostraca/Amphipoda		
Hyalellidae	*Hyalella azteca*	1
Insects		
Ephemeroptera		
Baetidae	*Baetis*	78
Isonychiidae	*Isonychia*	7
Heptageniidae	*Stenacron*	5
Heptageniidae	*Stenonema*	219
Tricorythidae	*Tricorythodes*	20
Odonata		
Calopterygidae	*Calopteryx*	3
Coenagrionidae	*Argia*	13
Gomphidae	juveniles	5
Hemiptera		
Corixidae		1
Belastomatidae	*Belastoma*	1
Trichoptera		
Hydropsychidae	*Ceratopsyche*	10
Hydropsychidae	*Hydropsyche*	20
Hydropsychidae	*Cheumatopsyche*	88
Megaloptera		
Corydalidae	*Corydalus*	1
Coleoptera		
Elmidae	*Stenelmis*	39
Diptera		
Chironomidae		49
Tipulidae	*Tipula*	1
	total abundance	629
	total richness	24
	EPT richness	8

<u>Caged Transplanted Mussel Study</u>

A dense population of plain pocketbooks, *Lampsilis cardium,* was located ~5 miles upstream of the Nature Preserve in Sugar Creek, ~6 miles west of McLean, IL, on Rte. 136, and organisms were transplanted to the Nature Preserve area. Mean weight, length, depth and TCI measurements are shown in table 12. Note that the TCI values for this species are much higher than the averages for those used in other locations. This is a function of the morphology of that particular species and not the area in which they were located.

Table 12. Size and condition measurements for caged plain pocketbooks, *Lampsilis cardium*, in Sugar Creek, Sandra Miller Bellrose Nature Preserve drainage, Logan Co., IL.

Site	Weight (g)	Length (mm)	Depth (mm)	TCI*
Sugar Creek (n = 5)	324.8 ± 33.6	123.1 ± 13.4	82.1 ± 13.7	8.08 ± 1.72

* TCI= tissue condition index, measured as dry weight of soft tissue divided by dry weight of shell, multiplied by 100.

Generally, elemental concentrations in dried mussel tissues were low, with two notable exceptions (Table 13); lead and zinc in these mussels were higher than at any other location in the entire study.

Table 13. Elemental concentrations in tissues (mg/kg dry weight) of plain pocketbooks (*Lampsilis cardium*, Unionidae) transplanted in Sugar Creek, Sandra Miller Bellrose Nature Preserve, Logan Co., IL

element	units	Sugar Ck average (n = 5)
Al	mg/kg	184
As	mg/kg	bdl
Ba	mg/kg	297
Ca	mg/kg	51,737
Cd	mg/kg	0.85
Co	mg/kg	1.1
Cr	mg/kg	1
Cu	mg/kg	5.3
Fe	mg/kg	865
Mg	mg/kg	2272
Mn	mg/kg	5860
Na	mg/kg	1,592
Ni	mg/kg	4.5
P	mg/kg	33,109
Pb	mg/kg	13.4
Se	mg/kg	7.225
Sr	mg/kg	102
Zn	mg/kg	688.5

Data for the last three locations (Little Wabash, Kaskaskia, and Sugar Creek) were generated for comparison upon conducting a similar study after discharges were put in place, so conclusions are not relevant at this time.

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APPENDIX B – EXHIBIT 3

Black Beauty Coal
Vermilion Grove Mine
Surface Water Quality Analysis

Prepared by
Peabody Energy
November 2010

The purpose of this report is to summarize surface water quality sampling results from Black Beauty Coal Company's Vermillion Grove Mine. Vermilion Grove Mine is an underground coal mine in east-central Illinois located upstream of the Little Vermilion River. The permitted area includes 411.5 acres that includes: a preparation plant, soil and coal stockpiles, rail road loop and rail load-out facility, ventilation shafts, office, shop, bath house, roads, refuse disposal area, diversions, and three sedimentation ponds in series. Runoff from the mine site is directed to sediment basin 003 (13SW-12), which discharges to an Unnamed Tributary of the Little Vermilion River.

As required by the NPDES permit dated January 10, 2001, the mine conducted sampling at the sedimentation pond Outfall 003 (13SW-12), at a sample location upstream (13SW-13) of the confluence of the unnamed tributary and Outfall 003, from upstream (11SW-14) and downstream (11SW-15) locations from the confluence of the unnamed tributary and the Little Vermilion River, and on the Little Vermilion River (Georgetown Lake) immediately above the Georgetown dam (6SW-16). Surface water sampling sites are summarized in Table 1 and shown on Figure 1. Samples were collected by grab sample methods during discharge events starting in 2001 and ending in 2007 for the five sample locations. Sample locations 13SW-12 and 13SW-13 include sample data into 2010. Up to a maximum of 10 discharge events per year were required to be sampled.

Permit requirements allow offsite discharge only when the flow rate in the receiving stream is three times that of the sediment basin outfall. Discharges from sedimentation pond Outfall 3 are controlled by a valve-structure. Samples from the surface water sites and the sedimentation basin discharge were analyzed for Temperature, DO, SpC, Volatile Suspended Solids, TSS, Total Ammonia, Alkalinity, Acidity, Hardness, pH, TDS, Cl, SO_4, Hg_T, Ba_T, Ba_D, B_T, B_D, Cd_T, Cd_D, $Cr(III)_T$, $Cr(III)_D$, Cu_T, Cu_D, Fe_T, Fe_D, Pb_T, Pb_D, Mn_T, Mn_D, Ni_T, Ni_D, Ag_T, Ag_D, Zn_T, and Zn_D. The data was analyzed using a variety of statistics including a comparison of the mean, maximum, and minimum chemical concentrations, Time series plots of individual parameters, and using an ANOVA statistic to compare population means of individual parameters between the sample points.

Tables 2 and 3 compare mean, maximum, and minimum chemical concentrations, respectively, of the mine outfall (13SW-12) to upstream sites (13SW-13 and 11SW-14). It can be seen that there are very few differences in chemical composition of mine affected water and upstream waters. Concentrations of inorganic chemicals, specifically sulfate, chloride, and TDS are higher at the mine outfall than in receiving streams. The higher concentrations of these parameters are likely due to the weathering of coal and refuse material contained within the mine site. Results for heavy metals indicate there is little or no difference between mine affected water and upstream waters.

Time series graphs of heavy metal concentrations at surface water sites are shown in Appendix B. Again, the graphs show concentrations at the mine outfall appear to be consistent with the up stream surface water stream sampling locations.

In order to determine if there are any statistically significance differences in mean concentrations of the heavy metals between the sample locations, an analysis of variance (ANOVA) was conducted using data from the mine outfall and the two upstream sites. According to EPA's Statistical Analysis of Ground-Water Monitoring Data at RCRA Facilities Unified Guidance March 2009, the analysis of variance is an acceptable method to use to compare mean concentrations. This procedure compares the means of different sampling locations and determines whether there are any significant differences among the sampling locations. Results of the ANOVA can be seen in Table 4. Based on this analysis, none of the heavy metals showed a statistically significant difference between the outfall mean concentration and the upstream mean concentration except for boron (dissolved and total), iron (total), and manganese (total). The ANOVA comparisons do not identify whether the outfall concentrations are statistically greater than the upstream locations only that there a statistically significant differences in mean concentrations.

Time series graphs of all chemicals that showed a statistically significant difference are shown in Appendix A. Time series graphs of all other heavy metals are shown in Appendix B.

As shown in Table 3, total boron concentrations are within the range of background and dissolved boron concentrations are comparable to background. Therefore, although the *mean* boron concentration shows a statistically significant difference, outfall concentrations are still comparable to the *range* of concentrations at upstream sampling sites. Furthermore, since mining has stopped and reclamation completed, boron concentrations have decreased to pre-mining levels. Total iron shows a similar relationship. The iron ranges seen during mining are within the range of background and again, since mining has stopped and reclamation completed, iron concentrations have decreased to pre-mining levels. Total manganese does show concentrations above those found in background and recent samples show that levels have not yet decreased to pre-mining concentrations. However, it is expected that manganese, similar to iron and boron, will return to pre-mining levels in time.

Under alkaline or neutral conditions, heavy metals do not readily leach out of coal or refuse materials and are not expected to be a significant component of mine runoff. For this reason, many materials handling processes are aimed specifically at minimizing any potential for acidic conditions to develop. These include minimizing stockpile area, minimizing exposure of disturbed areas and refuse, special handling requirements for coarse and fine refuse, and compaction and covering of material within the refuse pile. Furthermore, all upstream unaffected water is diverted around the mine site to prevent exposure to disturbed material and avoid unnecessary treatment of unaffected water.

In summary, Vermilion Grove Mine conducted surface water sampling both upstream and downstream of a sediment basin outfall that received all runoff from the mine site. After five years of sampling, this analysis has shown that, for the majority of analytes, concentrations at the mine outfall are not statistically different from background concentrations. Rather than requiring analysis of such an extensive list of analytes, the same level of protection could have been achieved through the use of indicator parameters.

TABLES

Table 1
Surface Water Monitoring Locations and Descriptions.

Vermilion Grove Site No.	Sample Point ID	Vermilion Grove Site Description	Sampling Date Range	No. of Sample Events
Site 1	13SW-12	Basin 003 Outfall	02/12/2001 - 07/26/2010	169
Site 2	13SW-13	Unnamed Tributary of LVR, upstream from 003 discharge	02/12/2001 - 07/26/2010	126
Site 3	11SW-14	Little Vermilion River, upstream from unnamed tributary	02/12/2001 - 04/04/2007	45
Site 4	11SW-15	Little Vermilion River, downstream from unnamed tributary	02/12/2001 - 04/04/2007	45
Site 5	6SW-16	Little Vermilion River, Georgetown Reservoir dam	02/12/2001 - 04/04/2007	45

Note: Table 1 includes sampling date range and total number of samples taken during the review period.

Table 2
Mean Chemical Concentrations
Vermilion Grove Sample Locations Analysis

Parameter	Units	13SW-12	13SW-13	11SW-14
		Average	Average	Average
Temp	[C°]	13.88	11.37	10.41
Hardness	[mg/L]	320	242	248
TDS	[mg/L]	949	404	368
pH Field	[S.U.]	8.05	7.93	7.90
pH Lab	[S.U.]	7.94	7.78	--
Acidity	[mg/L]	6	12	13
Alkalinity	[mg/L]	135	177	205
Cl	[mg/L]	244	78	51
SO4	[mg/L]	302	42	34
TSS	[mg/L]	25.40	34.23	70.23
SS	[mL/L]	0.17	--	--
DO	[mg/L]	9.46	9.35	8.97
Flow	[cfs]	10.63	47.07	147.78
Ba_D	[mg/L]	0.084	0.110	0.102
Ba_T	[mg/L]	0.055	0.057	0.062
B_D	[mg/L]	0.142	0.077	0.067
B_T	[mg/L]	0.144	0.085	0.070
Cd_D	[mg/L]	0.002	0.002	0.002
Cd_T	[mg/L]	0.002	0.002	0.002
Cr_D	[mg/L]	0.002	0.002	0.002
Cr_T	[mg/L]	0.003	0.004	0.004
Cu_D	[mg/L]	0.005	0.003	0.004
Cu_T	[mg/L]	0.003	0.004	0.005
Fe_D	[mg/L]	0.093	0.162	0.177
Fe_T	[mg/L]	0.946	1.981	2.335
Pb_D	[mg/L]	0.002	0.002	0.002
Pb_T	[mg/L]	0.002	0.002	0.002
Mn_D	[mg/L]	0.086	0.044	0.022
Mn_T	[mg/L]	0.211	0.065	0.060
Ni_D	[mg/L]	0.005	0.003	0.002
Ni_T	[mg/L]	0.005	0.004	0.003
Ag_D	[mg/L]	0.002	0.002	0.002
Ag_T	[mg/L]	0.002	0.002	0.002
Zn_D	[mg/L]	0.027	0.033	0.030
Zn_T	[mg/L]	0.014	0.021	0.021
Hg_D	[mg/L]	0.0002	0.0002	0.0002
Hg_T	[mg/L]	0.0002	0.0002	0.0002
Cr(VI)	[mg/L]	0.018	0.018	0.018
Cr(III)	[mg/L]	0.010	0.012	0.012
Cr(IIID)	[mg/L]	0.004	0.003	0.003
NH_{4T}	[mg/L]	1.05	1.00	1.01

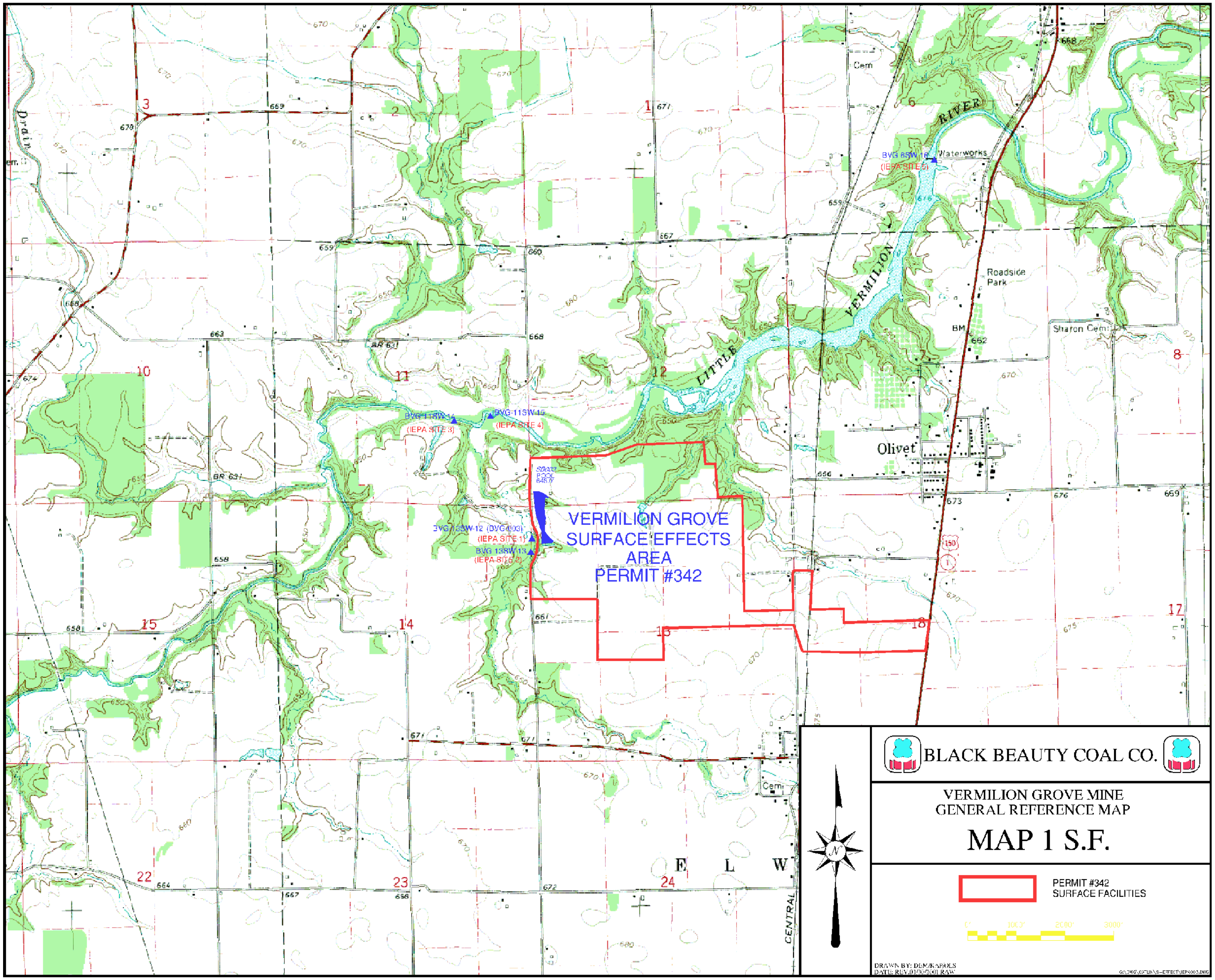
Table 3
Maximum and Minimum Chemical Concentrations
Vermilion Grove Sample Locations

Parameter	Units	13SW-12		13SW-13		11SW-14	
		Minimum	Maximum	Minimum	Maximum	Minimum	Maximum
Temp	[C°]	1.89	28.20	2.60	24.30	2.78	24.40
Hardness	[mg/L]	50	780	140	340	110	370
TDS	[mg/L]	166	4608	192	1277	134	870
pH Field	[S.U.]	6.42	8.82	6.44	8.60	6.37	9.30
pH Lab	[S.U.]	6.88	9.28	7.16	8.20	--	--
Acidity	[mg/L]	-74	123	1	164	1	122
Alkalinity	[mg/L]	52	330	74	288	85	408
Cl	[mg/L]	10	767	10	742	3	394
SO4	[mg/L]	10	935	10	308	1	148
TSS	[mg/L]	1.00	198	1.00	190	2.00	327
SS	[mL/L]	0.04	0.40	--	--	--	--
DO	[mg/L]	5.84	17.44	5.32	16.60	5.02	11.88
Flow	[cfs]	0.00	143.91	0.88	540.27	125.00	155.00
Ba_D	[mg/L]	0.031	0.320	0.027	0.544	0.030	0.864
Ba_T	[mg/L]	0.031	0.082	0.033	0.136	0.040	0.137
B_D	[mg/L]	0.041	0.294	0.020	0.281	0.015	0.218
B_T	[mg/L]	0.022	0.501	0.002	0.652	0.014	0.443
Cd_D	[mg/L]	0.002	0.002	0.002	0.002	0.002	0.002
Cd_T	[mg/L]	0.002	0.002	0.002	0.002	0.002	0.002
Cr_D	[mg/L]	0.002	0.004	0.002	0.008	0.002	0.004
Cr_T	[mg/L]	0.002	0.038	0.002	0.026	0.002	0.026
Cu_D	[mg/L]	0.002	0.087	0.002	0.038	0.002	0.030
Cu_T	[mg/L]	0.002	0.010	0.002	0.027	0.002	0.022
Fe_D	[mg/L]	0.005	0.889	0.005	1.160	0.005	1.640
Fe_T	[mg/L]	0.005	11.90	0.005	14.50	0.005	15.70
Pb_D	[mg/L]	0.002	0.005	0.002	0.003	0.002	0.005
Pb_T	[mg/L]	0.002	0.006	0.002	0.006	0.002	0.006
Mn_D	[mg/L]	0.002	1.22	0.002	0.848	0.004	0.190
Mn_T	[mg/L]	0.003	1.17	0.003	0.738	0.002	0.246
Ni_D	[mg/L]	0.002	0.050	0.002	0.033	0.002	0.004
Ni_T	[mg/L]	0.002	0.074	0.002	0.055	0.002	0.025
Ag_D	[mg/L]	0.002	0.002	0.002	0.002	0.002	0.006
Ag_T	[mg/L]	0.002	0.005	0.002	0.008	0.002	0.007
Zn_D	[mg/L]	0.002	0.134	0.002	0.152	0.002	0.157
Zn_T	[mg/L]	0.002	0.154	0.002	0.123	0.002	0.142
Hg_D	[mg/L]	0.0002	0.0002	0.0002	0.0002	0.0002	0.0002
Hg_T	[mg/L]	0.0002	0.0007	0.0002	0.0002	0.0002	0.0002
Cr(VI)	[mg/L]	0.010	0.020	0.010	0.020	0.010	0.020
Cr(III)	[mg/L]	0.002	0.038	0.002	0.026	0.002	0.026
Cr(IIID)	[mg/L]	0.002	0.020	0.002	0.020	0.002	0.020
NH_{4T}	[mg/L]	1.00	2.00	1.00	1.10	1.00	1.30

Table 4
Values Computed in ANOVA Statistical Analysis.

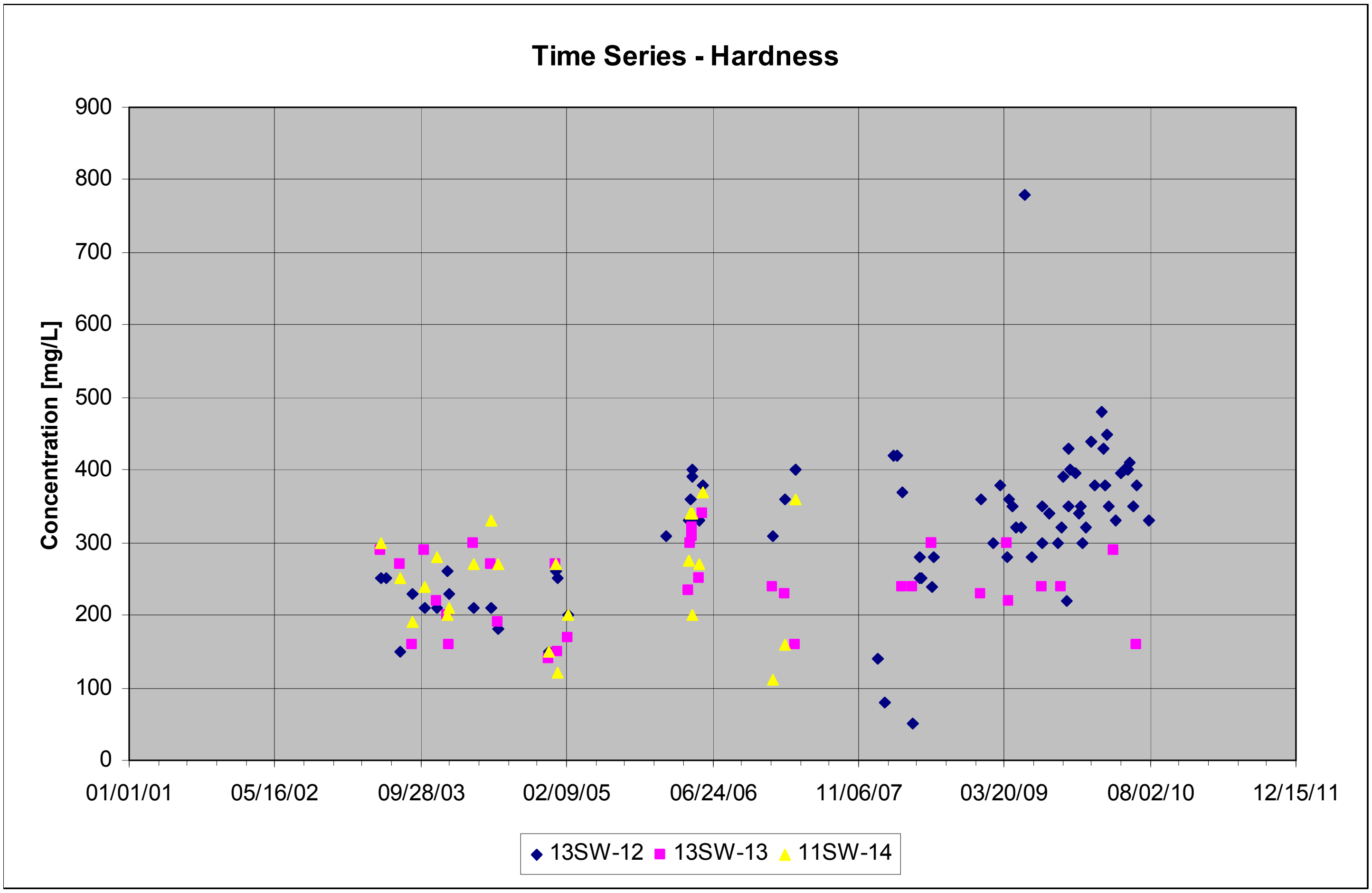
Parameter	Sum of Squares (Wells)	Sum of Squares (Total)	Sum of Squares (Error)	Mean Squares (Wells)	Mean Squares (Error)	Degrees of Freedom (p-1)	Degrees of Freedom (N-p)	F Value (Calculated)	F Value (Table)	Equal Means (All Wells)
Hardness	188106	1196439	1008333	94053	7756	2	130	12.13	3.07	Significant Difference (95%)
TDS	9390606	39623611	30233005	4695303	236195	2	128	19.88	3.07	Significant Difference (95%)
pH Field	0.97	39.59	38.62	0.49	0.20	2	196	2.47	3.07	No Significant Difference
Acidity	2210	136509	134298	1105	678	2	198	1.63	3.07	No Significant Difference
Alkalinity	168965	685086	516121	84483	2607	2	198	32.41	3.07	Significant Difference (95%)
Cl	1640219	6757352	5117134	820109	25208	2	203	32.53	3.07	Significant Difference (95%)
SO_4	3553664	9231666	5678002	1776832	27970	2	203	63.53	3.07	Significant Difference (95%)
TSS	62272	589382	527111	31136	2649	2	199	11.75	3.07	Significant Difference (95%)
DO	5.91	398.48	392.57	2.95	2.93	2	134	1.01	3.07	No Significant Difference
Ba_D	0.02	1.83	1.82	0.01	0.01	2	128	0.54	3.07	No Significant Difference
Ba_T	0.00	0.04	0.04	0.00	0.00	2	128	1.38	3.07	No Significant Difference
B_D	0.15	0.58	0.43	0.07	0.00	2	128	22.01	3.07	Significant Difference (95%)
B_T	0.14	1.13	0.99	0.07	0.01	2	128	8.76	3.07	Significant Difference (95%)
Cd_D	0.0000	0.0000	0.0000	0.0000	0.0000	2	128	0.00	3.07	No Significant Difference
Cd_T	0.0000	0.0000	0.0000	0.0000	0.0000	2	128	0.00	3.07	No Significant Difference
Cr_D	0.0000	0.0001	0.0001	0.0000	0.0000	2	127	0.32	3.07	No Significant Difference
Cr_T	0.0000	0.0030	0.0030	0.0000	0.0000	2	127	0.20	3.07	No Significant Difference
Cu_D	0.0000	0.0102	0.0102	0.0000	0.0001	2	128	0.19	3.07	No Significant Difference
Cu_T	0.0001	0.0024	0.0023	0.0000	0.0000	2	128	2.34	3.07	No Significant Difference
Fe_D	0.18	8.79	8.61	0.09	0.07	2	128	1.35	3.07	No Significant Difference
Fe_T	74.49	1418.59	1344.09	37.25	6.79	2	198	5.49	3.07	Significant Difference (95%)
Pb_D	0.0000	0.0000	0.0000	0.0000	0.0000	2	128	0.39	3.07	No Significant Difference
Pb_T	0.0000	0.0001	0.0001	0.0000	0.0000	2	128	0.30	3.07	No Significant Difference
Mn_D	0.10	3.56	3.46	0.05	0.03	2	128	1.76	3.07	No Significant Difference
Mn_T	1.08	9.36	8.29	0.54	0.04	2	195	12.64	3.07	Significant Difference (95%)
Ni_D	0.0001	0.0048	0.0047	0.0001	0.0000	2	128	1.86	3.07	No Significant Difference
Ni_T	0.0001	0.0099	0.0098	0.0000	0.0001	2	128	0.55	3.07	No Significant Difference
Ag_D	0.0000	0.0000	0.0000	0.0000	0.0000	2	128	1.02	3.07	No Significant Difference
Ag_T	0.0000	0.0001	0.0001	0.0000	0.0000	2	128	0.43	3.07	No Significant Difference
Zn_D	0.0009	0.1484	0.1475	0.0005	0.0012	2	128	0.39	3.07	No Significant Difference
Zn_T	0.0014	0.0896	0.0882	0.0007	0.0007	2	128	1.00	3.07	No Significant Difference
Hg_D	0.0000	0.0000	0.0000	0.0000	0.0000	2	127	0.00	3.07	No Significant Difference
Hg_T	0.0000	0.0000	0.0000	0.0000	0.0000	2	128	1.33	3.07	No Significant Difference
Cr(VI)	0.0000	0.0021	0.0021	0.0000	0.0000	2	127	0.00	3.07	No Significant Difference
Cr(III)	0.0001	0.0096	0.0095	0.0000	0.0001	2	127	0.55	3.07	No Significant Difference
Cr(IIID)	0.0000	0.0022	0.0022	0.0000	0.0000	2	127	0.19	3.07	No Significant Difference
NH_{4T}	0.0513	2.0680	2.0167	0.0256	0.0159	2	127	1.61	3.07	No Significant Difference

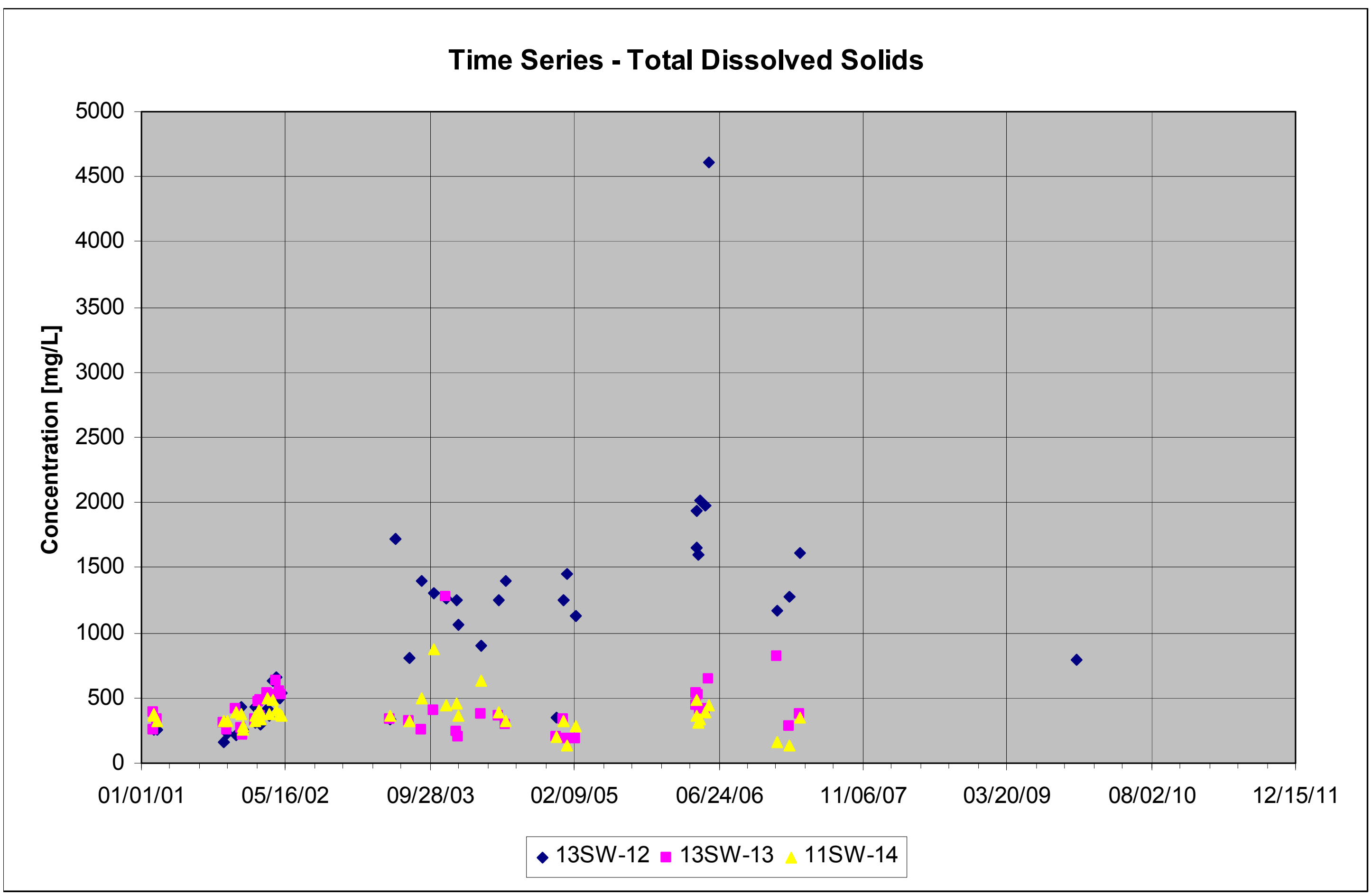
Figure 1
Vermilion Grove Mine Surface Water Sampling Locations.

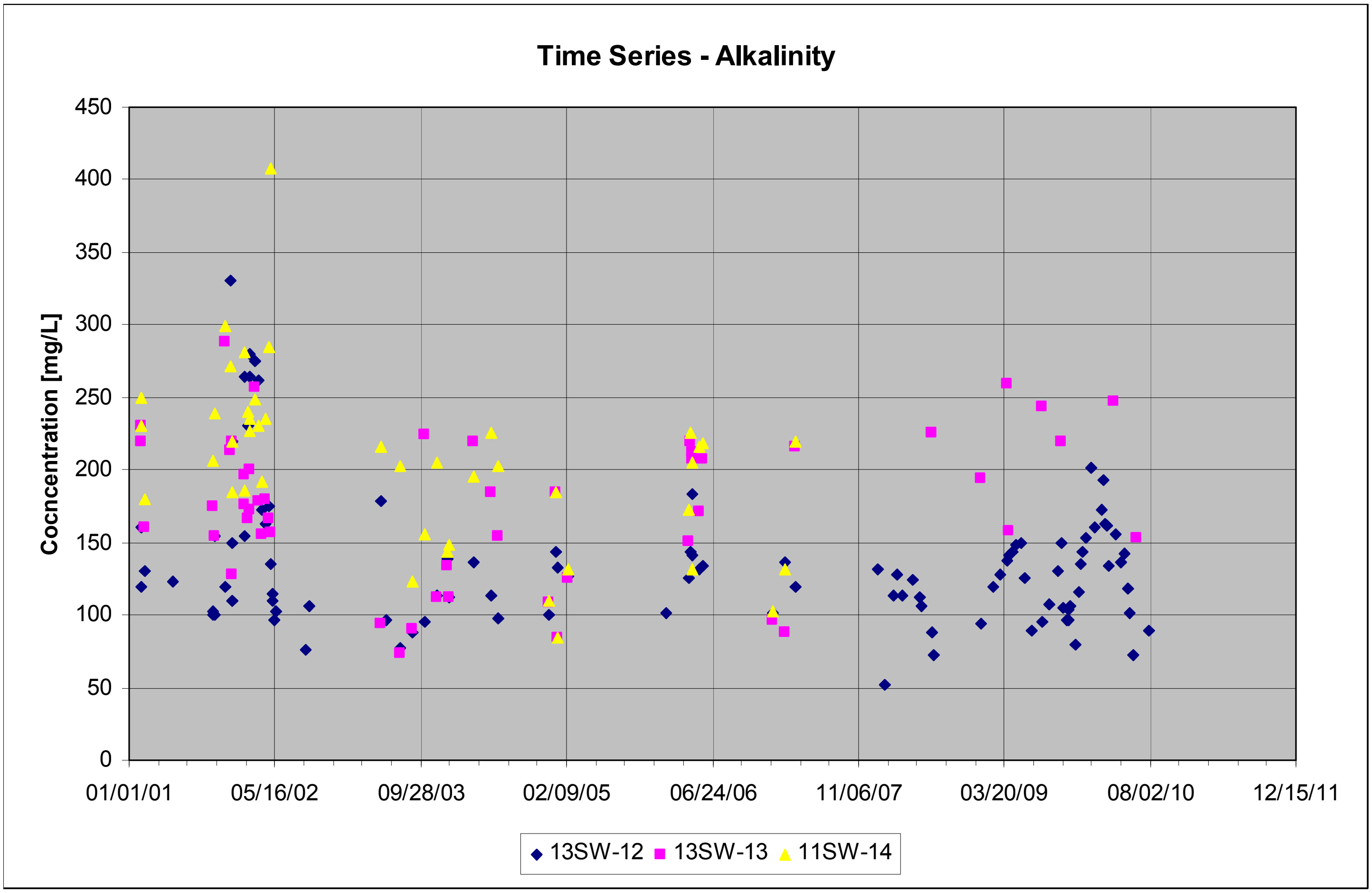


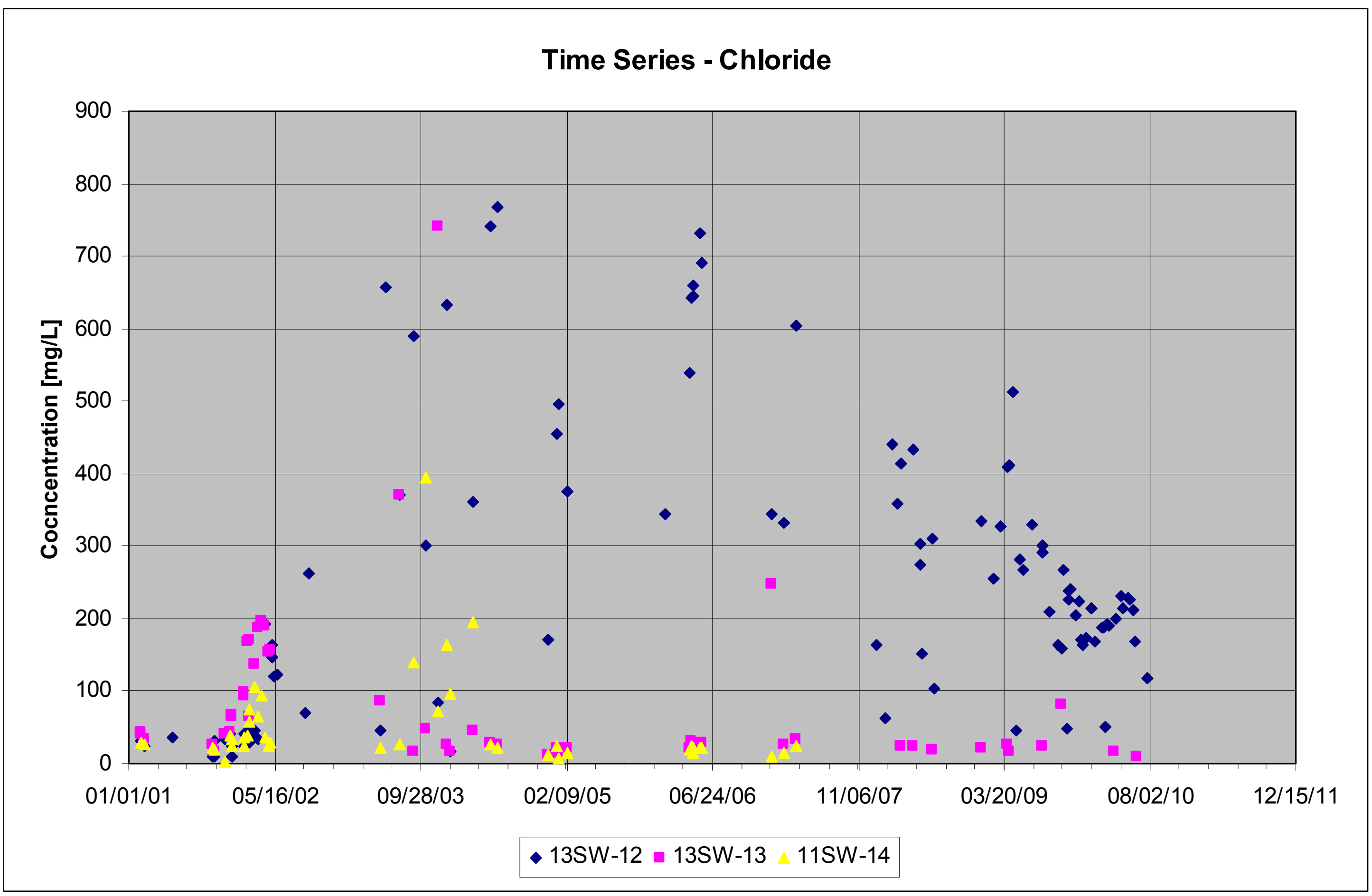
Appendix A

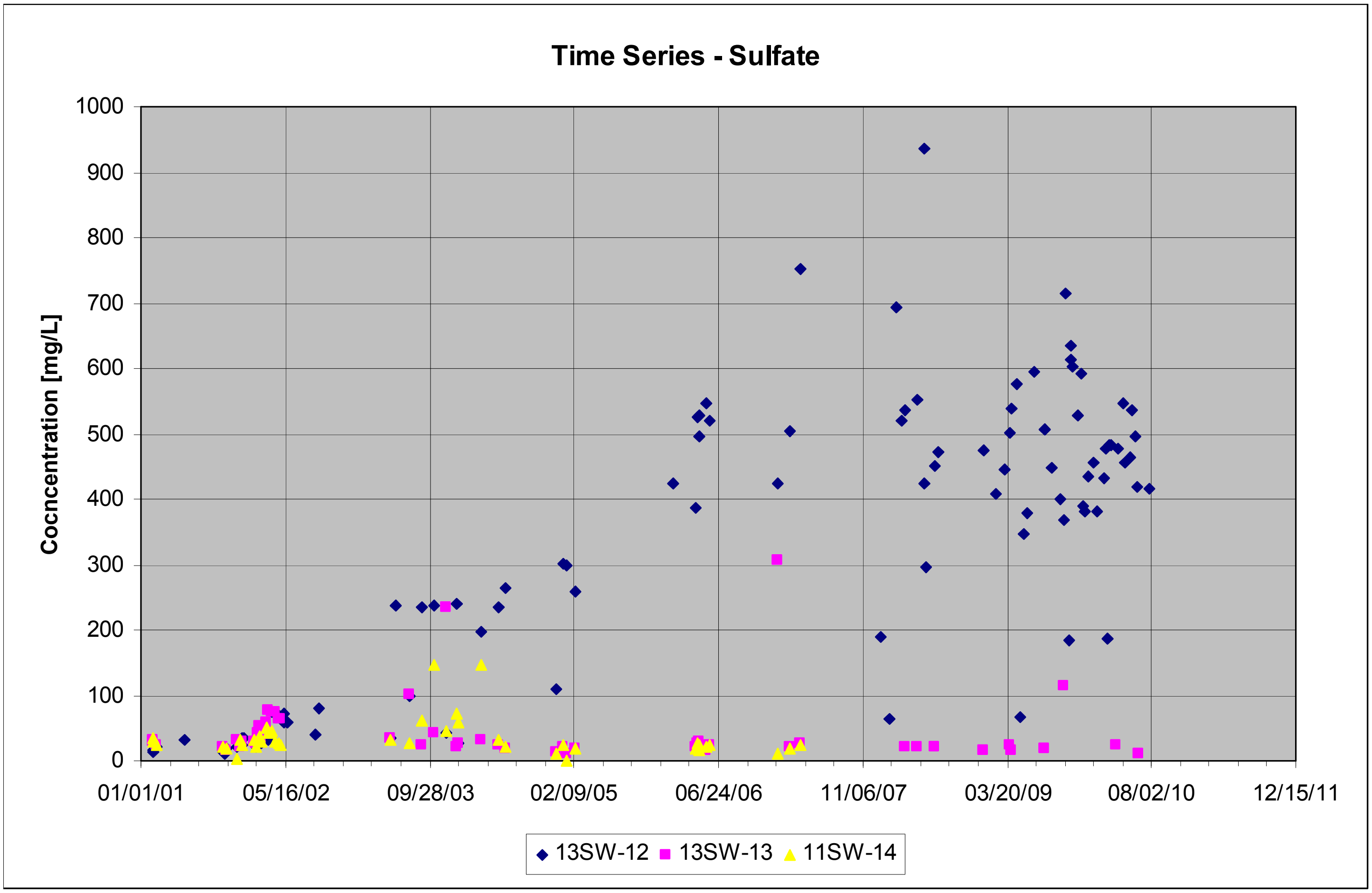
Time Series Graphs for Chemicals Showing A Statistically Significance Difference


Time Series - Hardness
Concentration [mg/L]
900
800
700
600
500
400
300
200
100
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

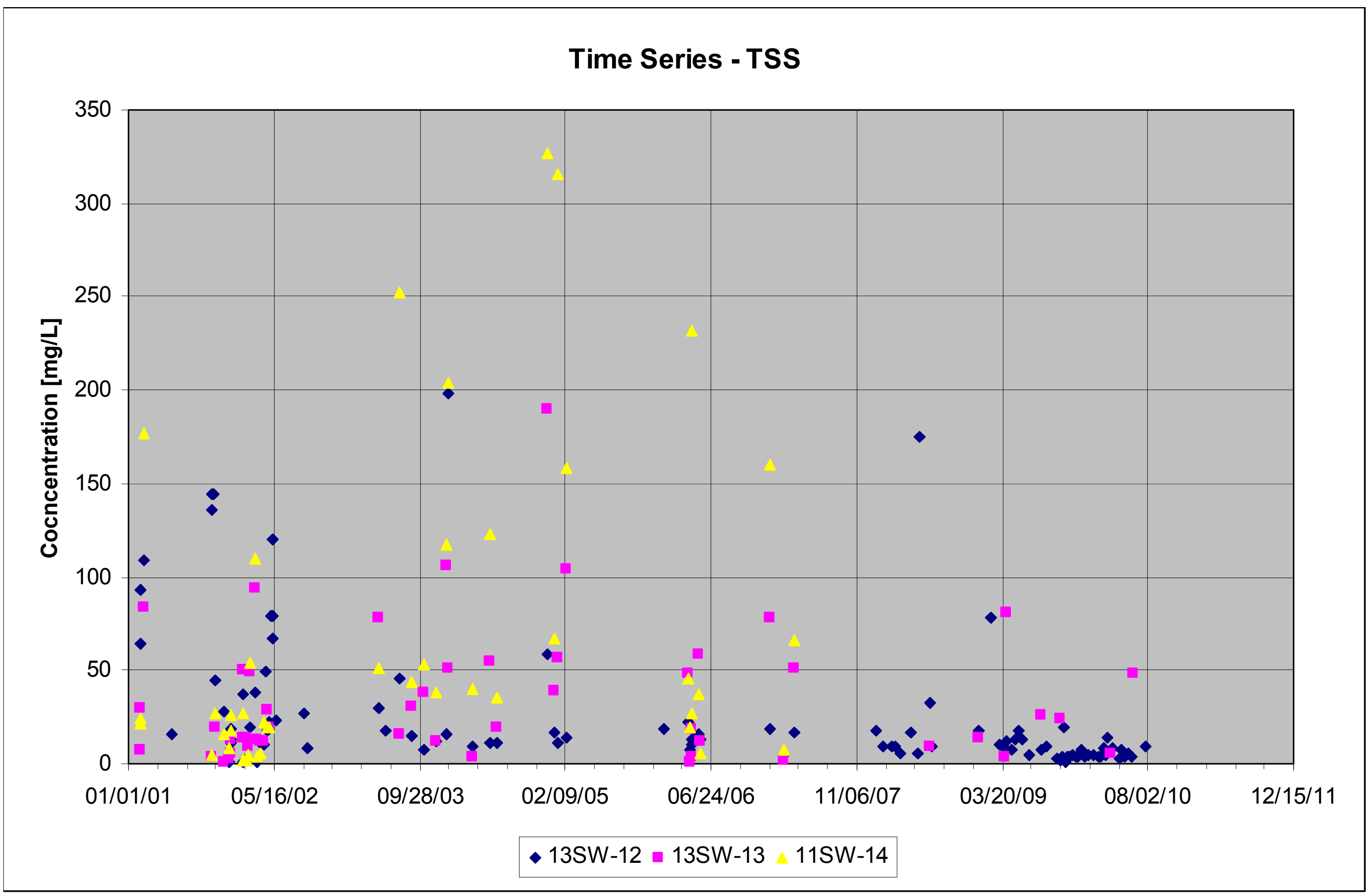

Time Series - Total Dissolved Solids
Concentration [mg/L]
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14


Time Series - Alkalinity
Cocncentration [mg/L]
450
400
350
300
250
200
150
100
50
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

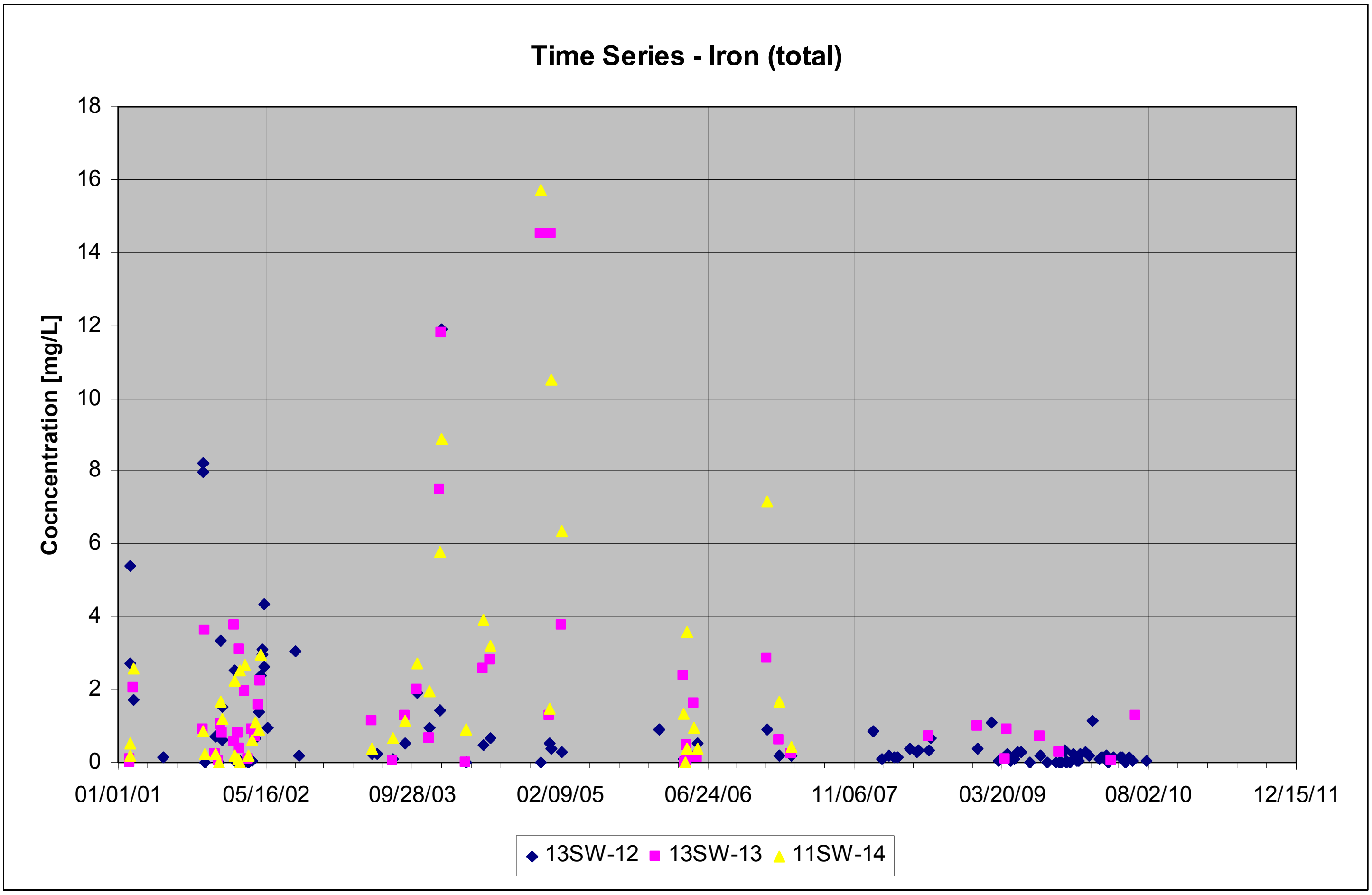

Time Series - Chloride
Cocncentration [mg/L]
900
800
700
600
500
400
300
200
100
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



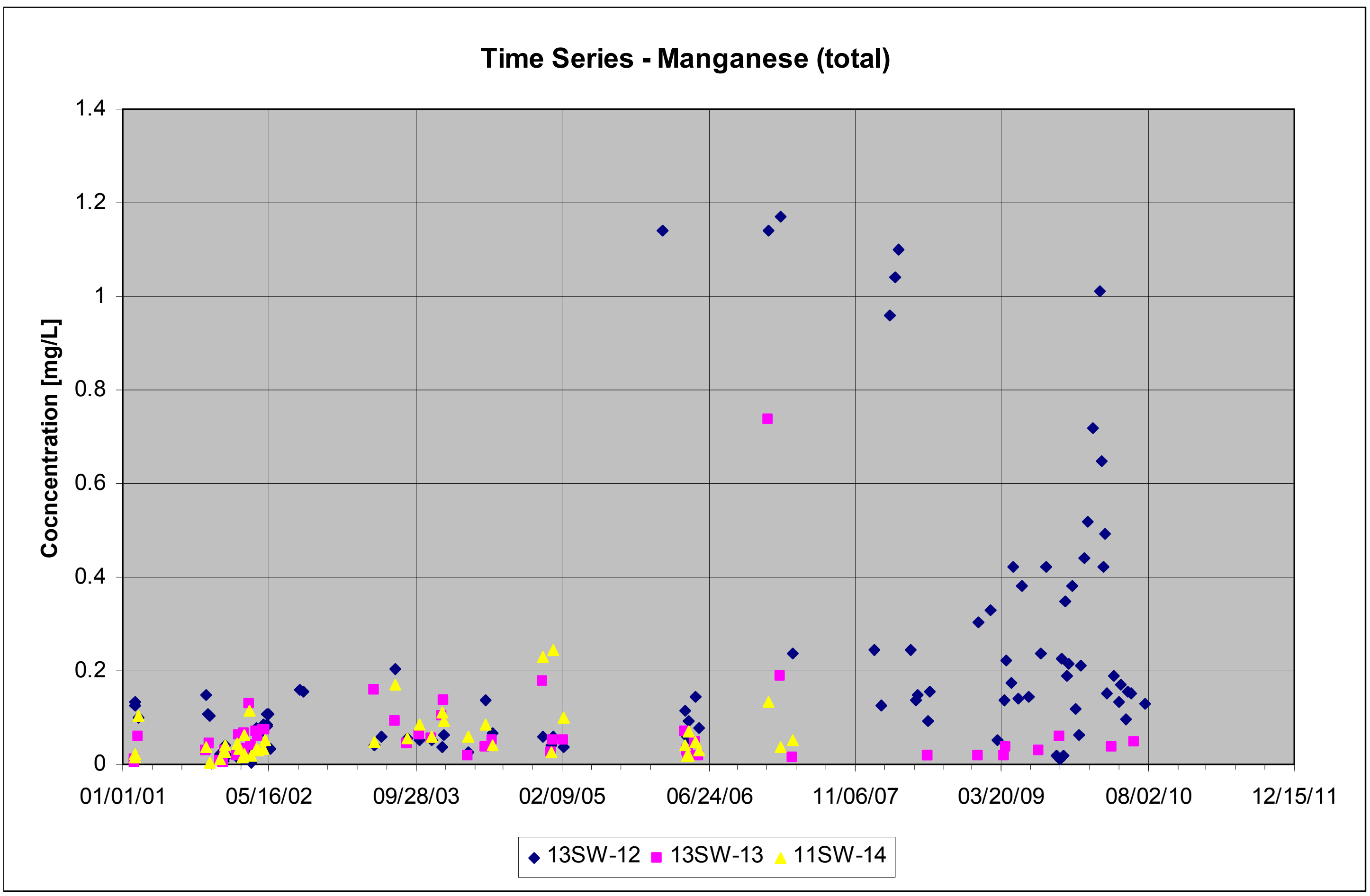
Time Series - Sulfate
1000
900
800
700
600
500
400
300
200
100
0
Cocncentration [mg/L]
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



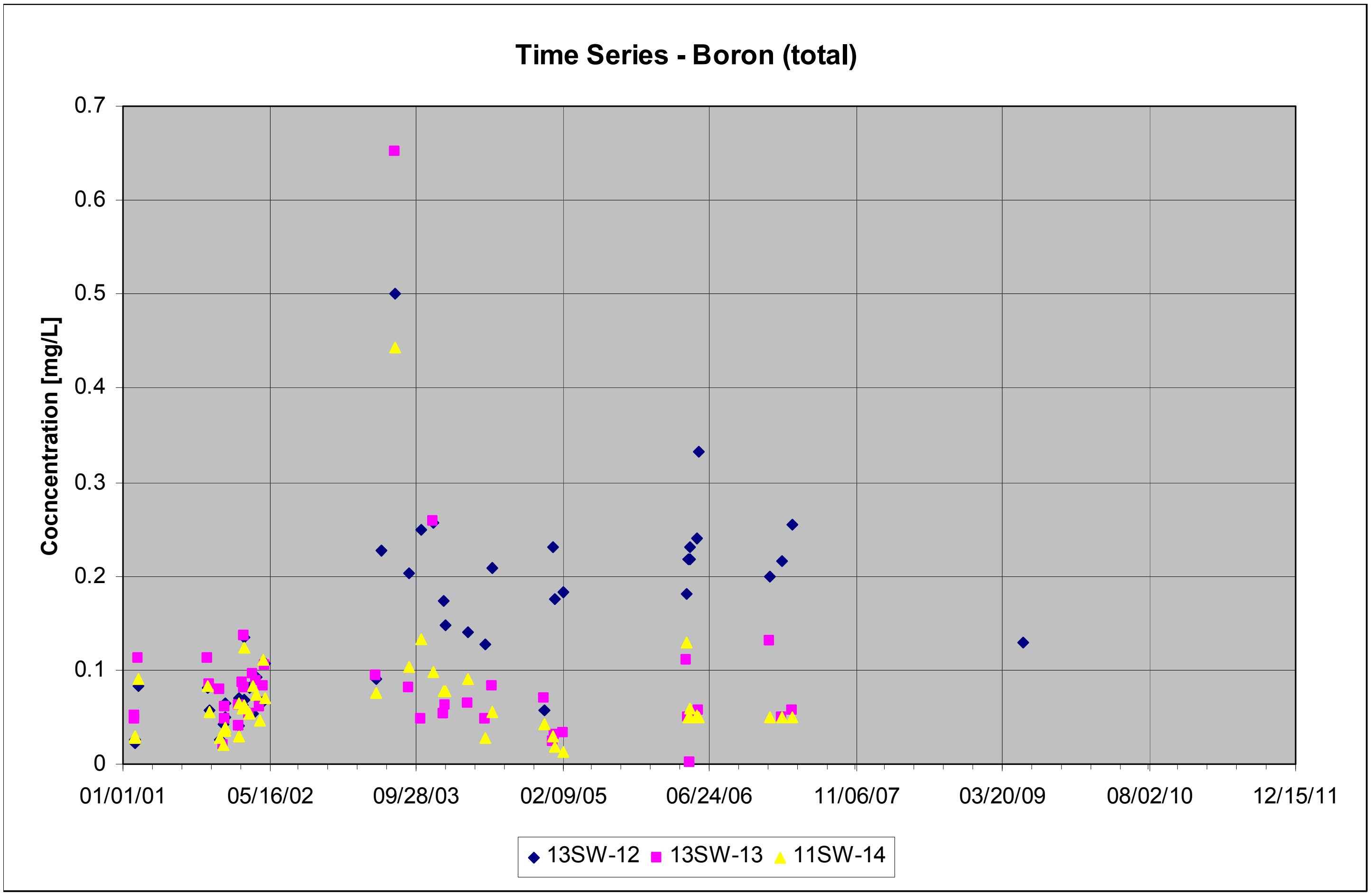
Time Series - TSS
350
300
250
200
150
100
50
0
Cocncentration [mg/L]
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



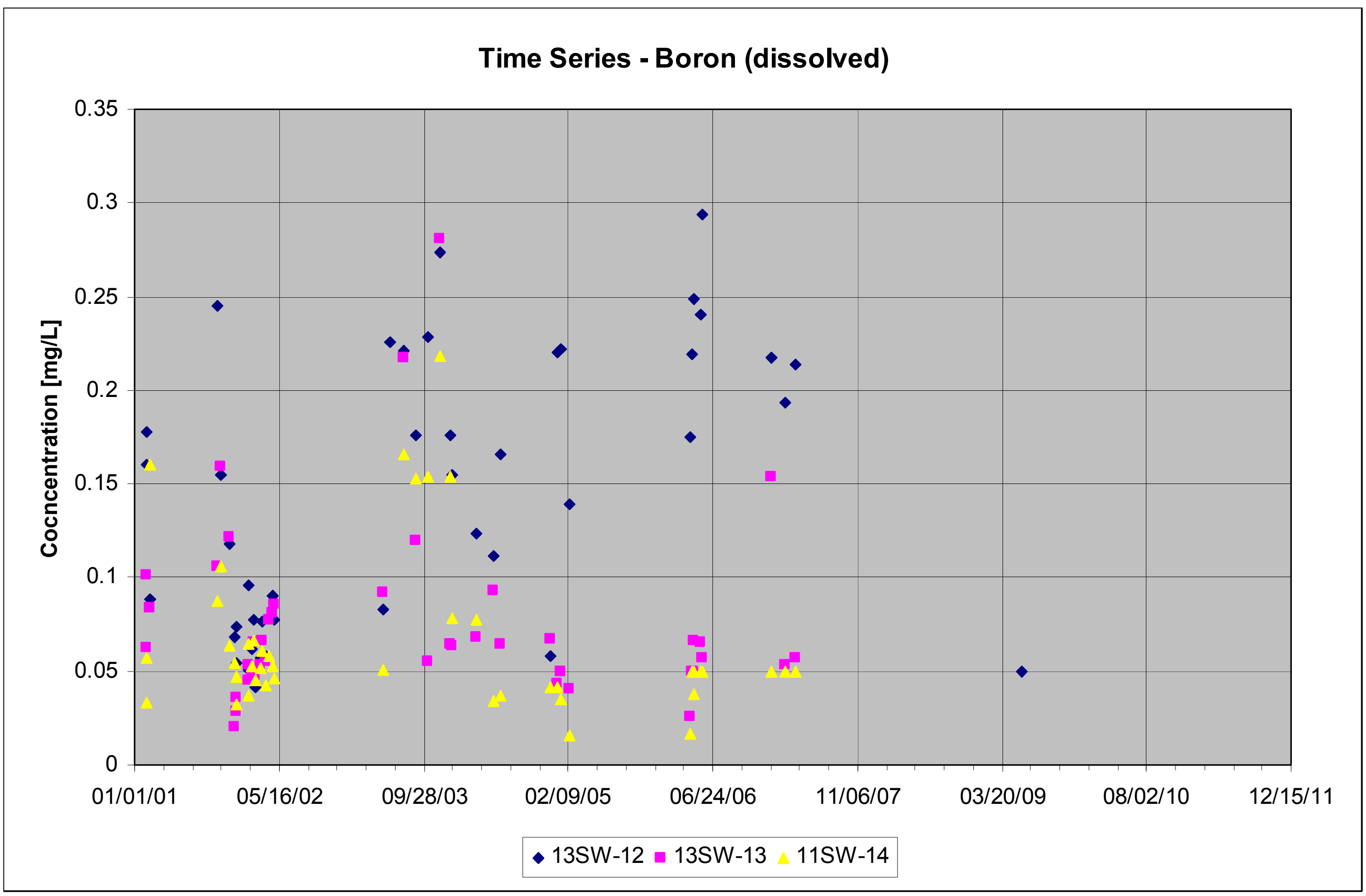
Time Series - Iron (total)
Cocncentration [mg/L]
0
2
4
6
8
10
12
14
16
18
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



Time Series - Manganese (total)
Cocncentration [mg/L]
0
0.2
0.4
0.6
0.8
1
1.2
1.4
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



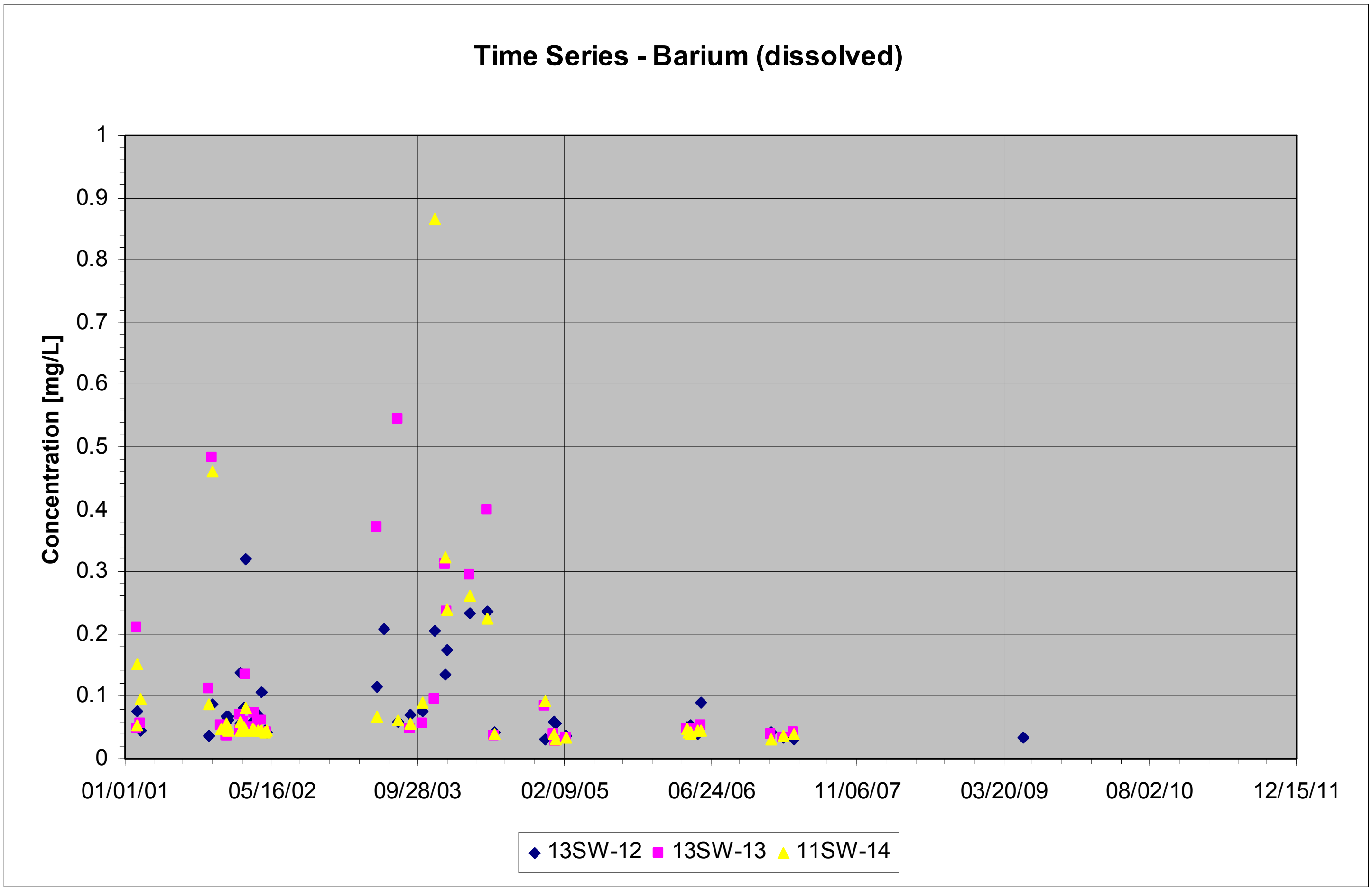
Time Series - Boron (total)
Cocncentration [mg/L]
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

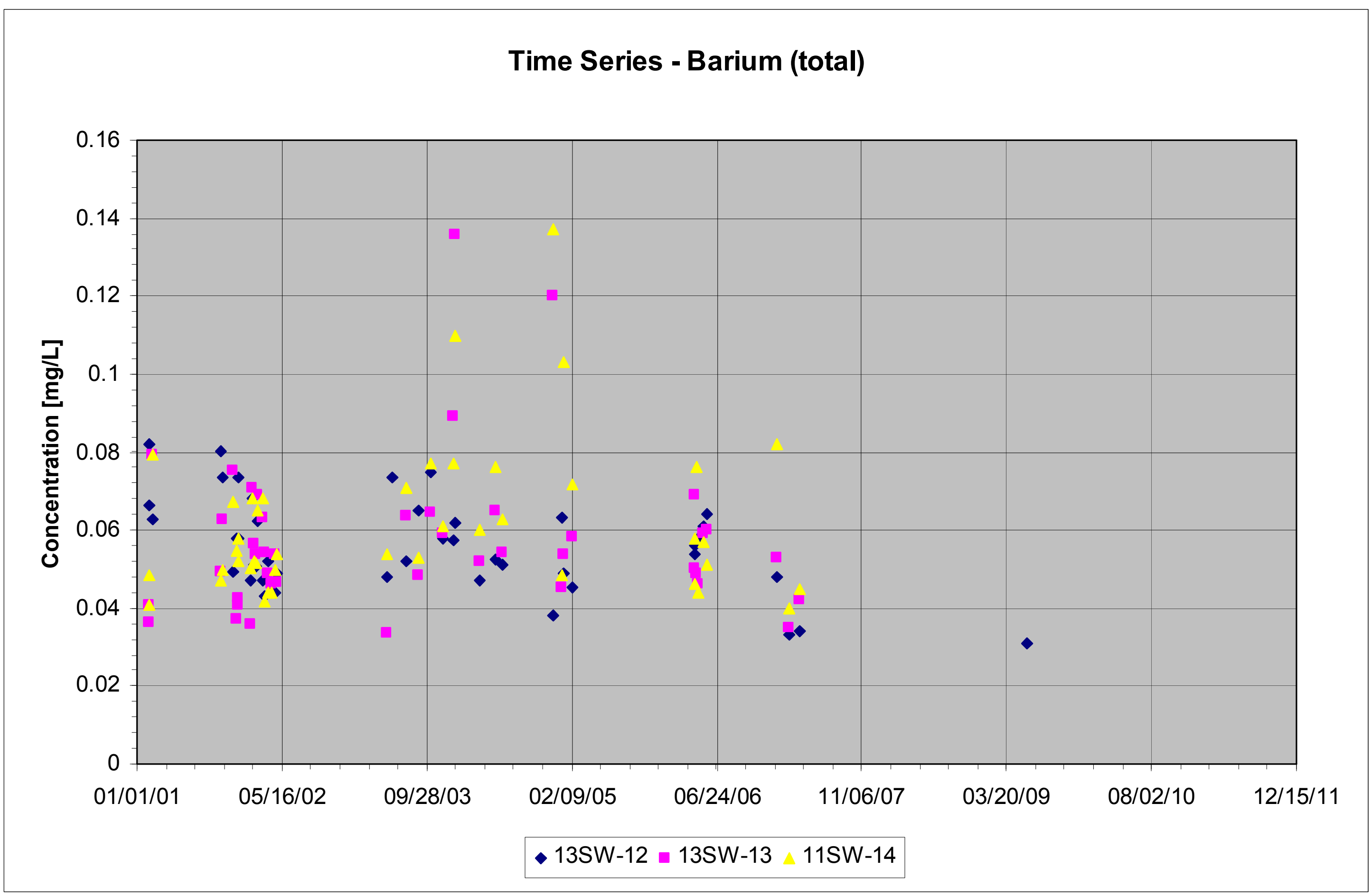


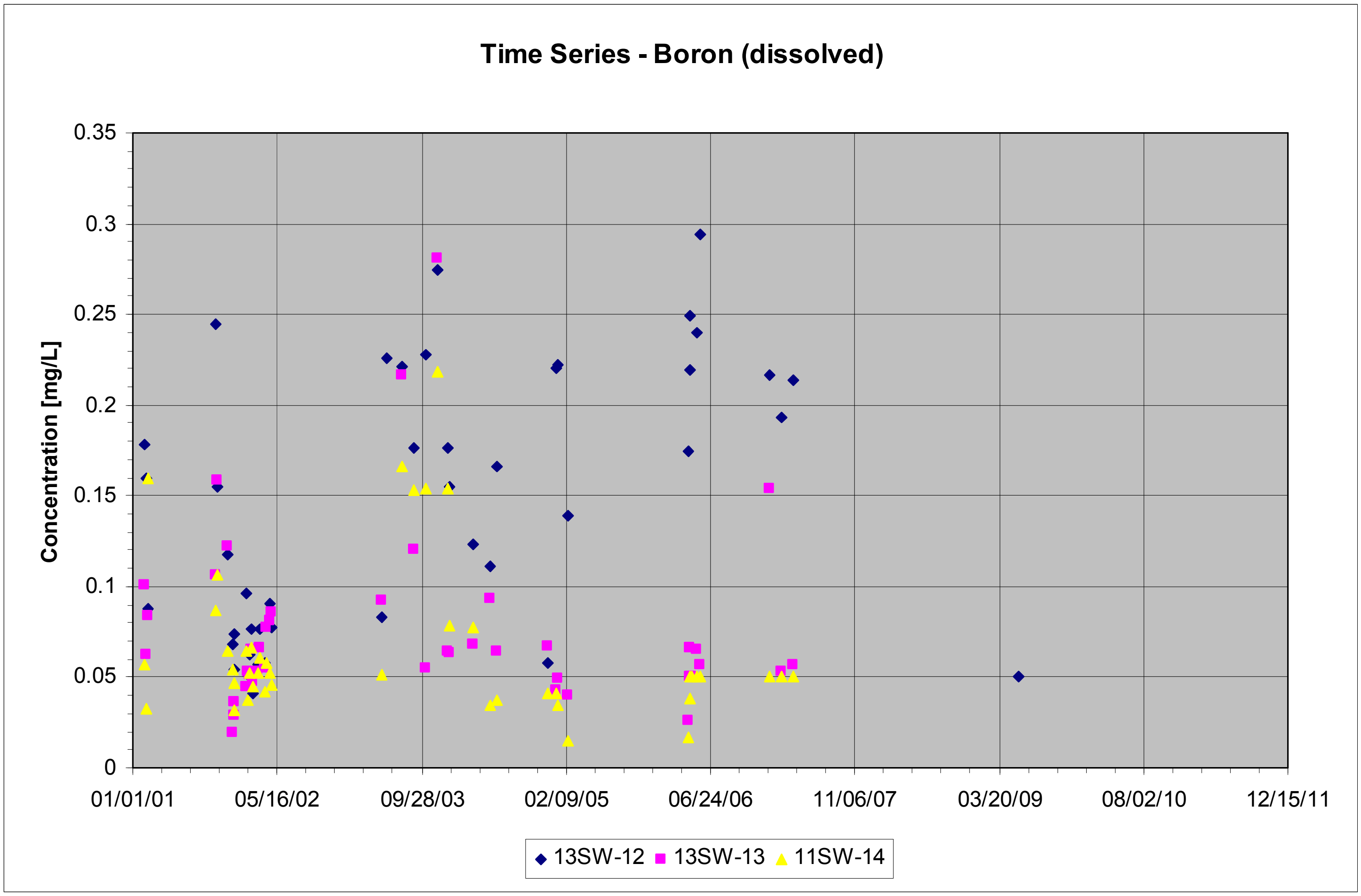
Time Series - Boron (dissolved)
Cocncentration [mg/L]
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

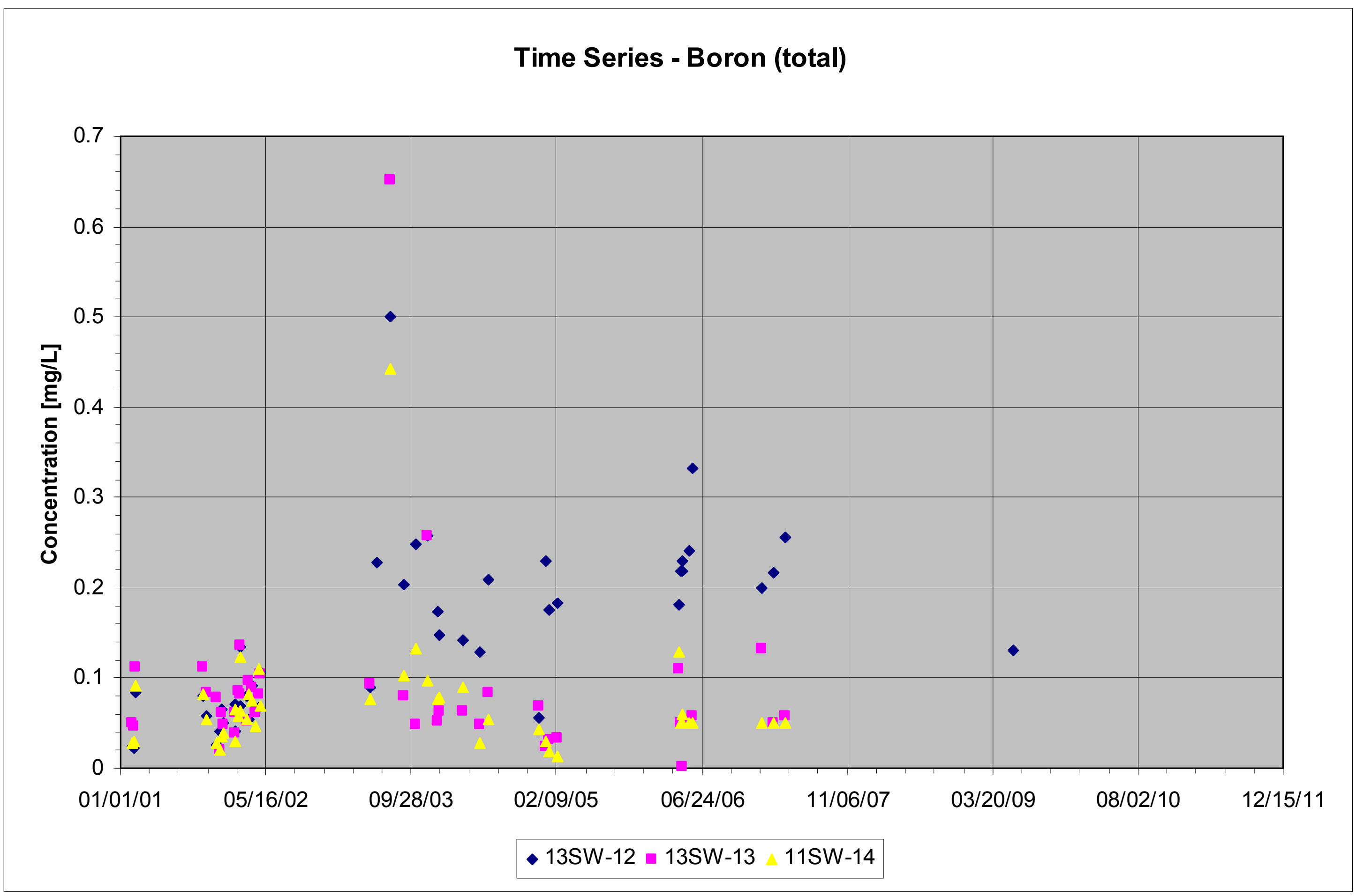
Appendix B

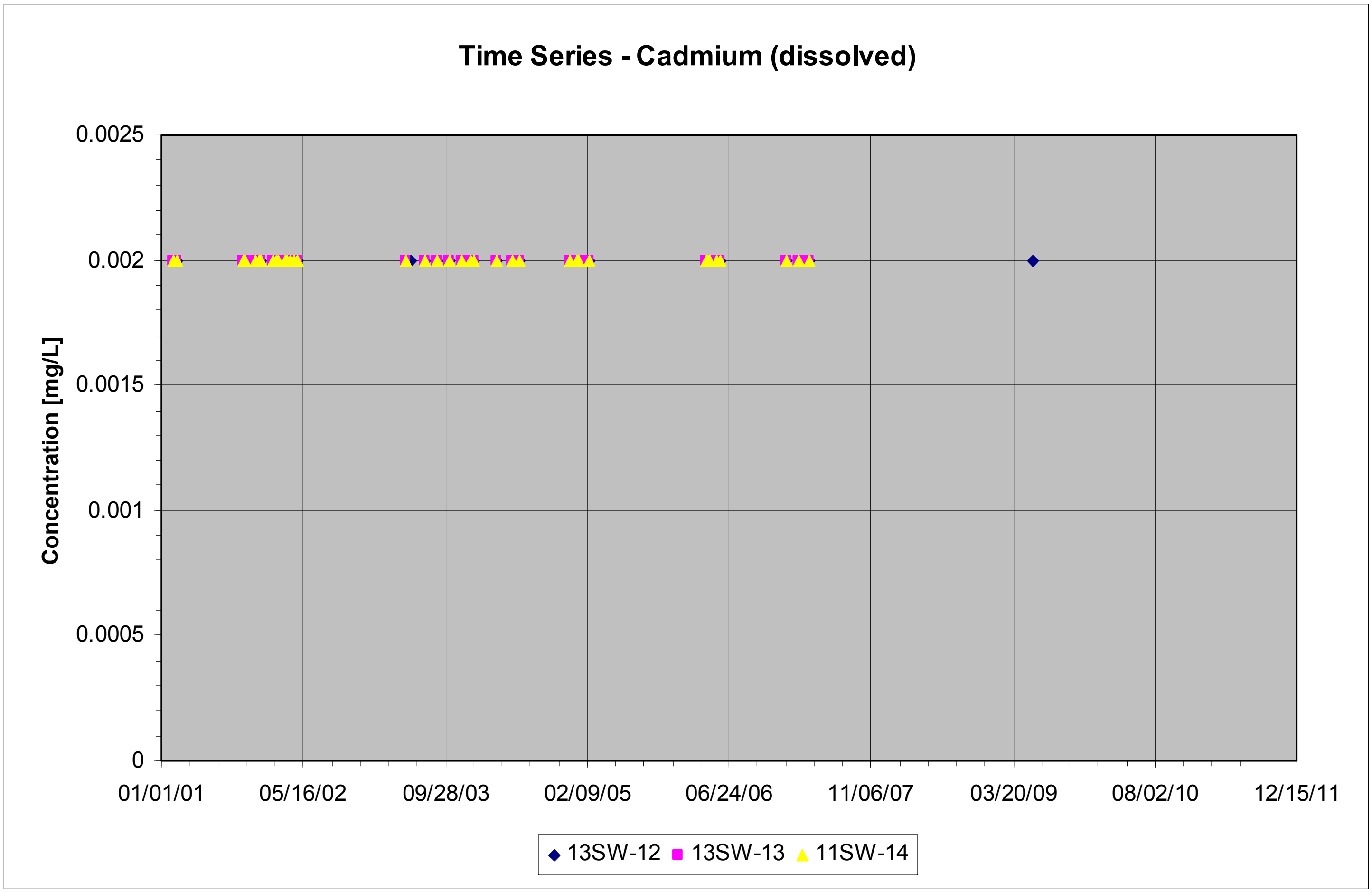
Time Series Graphs
Heavy Metals


Time Series - Barium (dissolved)
Concentration [mg/L]
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

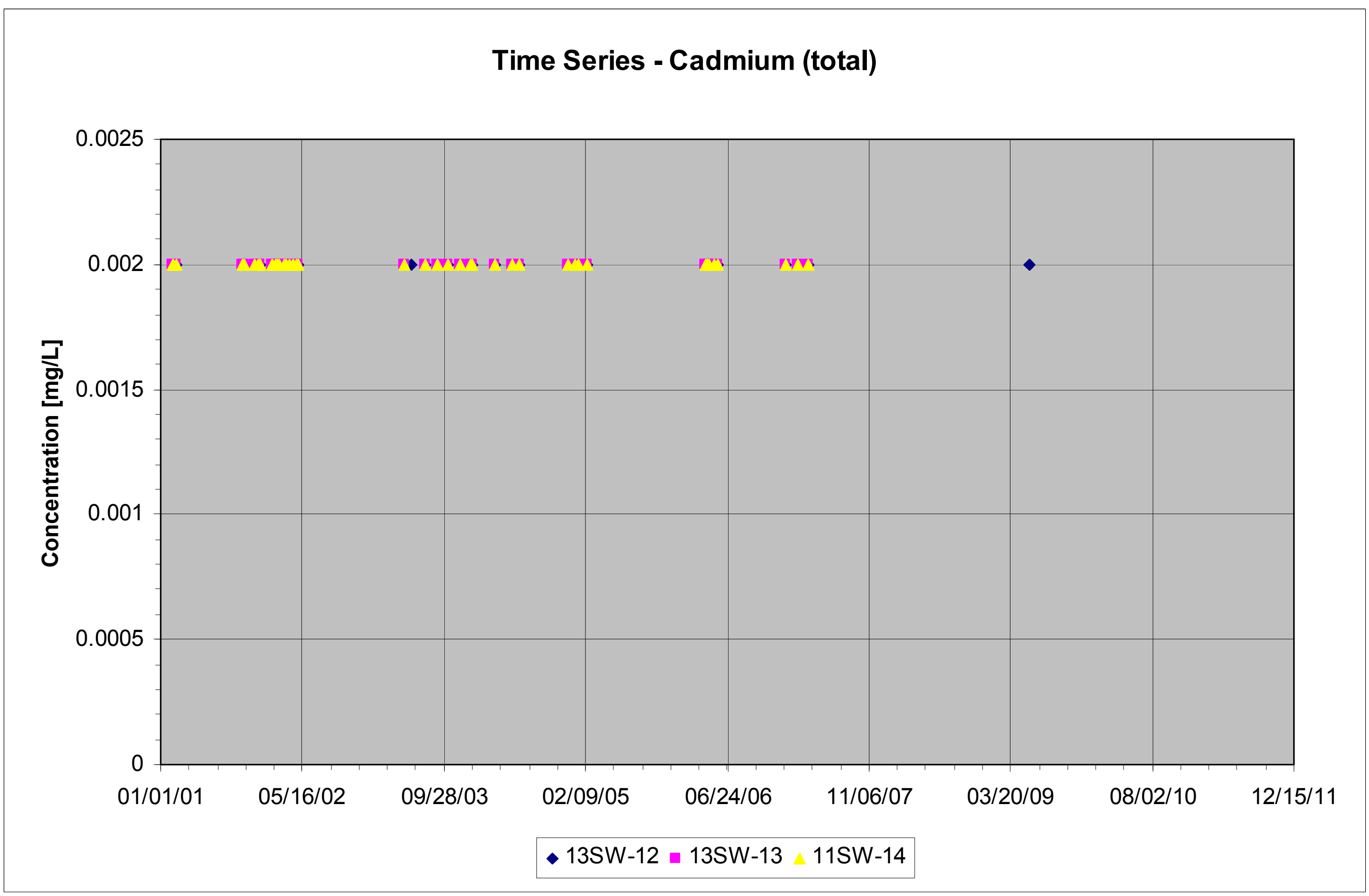

Time Series - Barium (total)
Concentration [mg/L]
0.16
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14


Time Series - Boron (dissolved)
Concentration [mg/L]
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

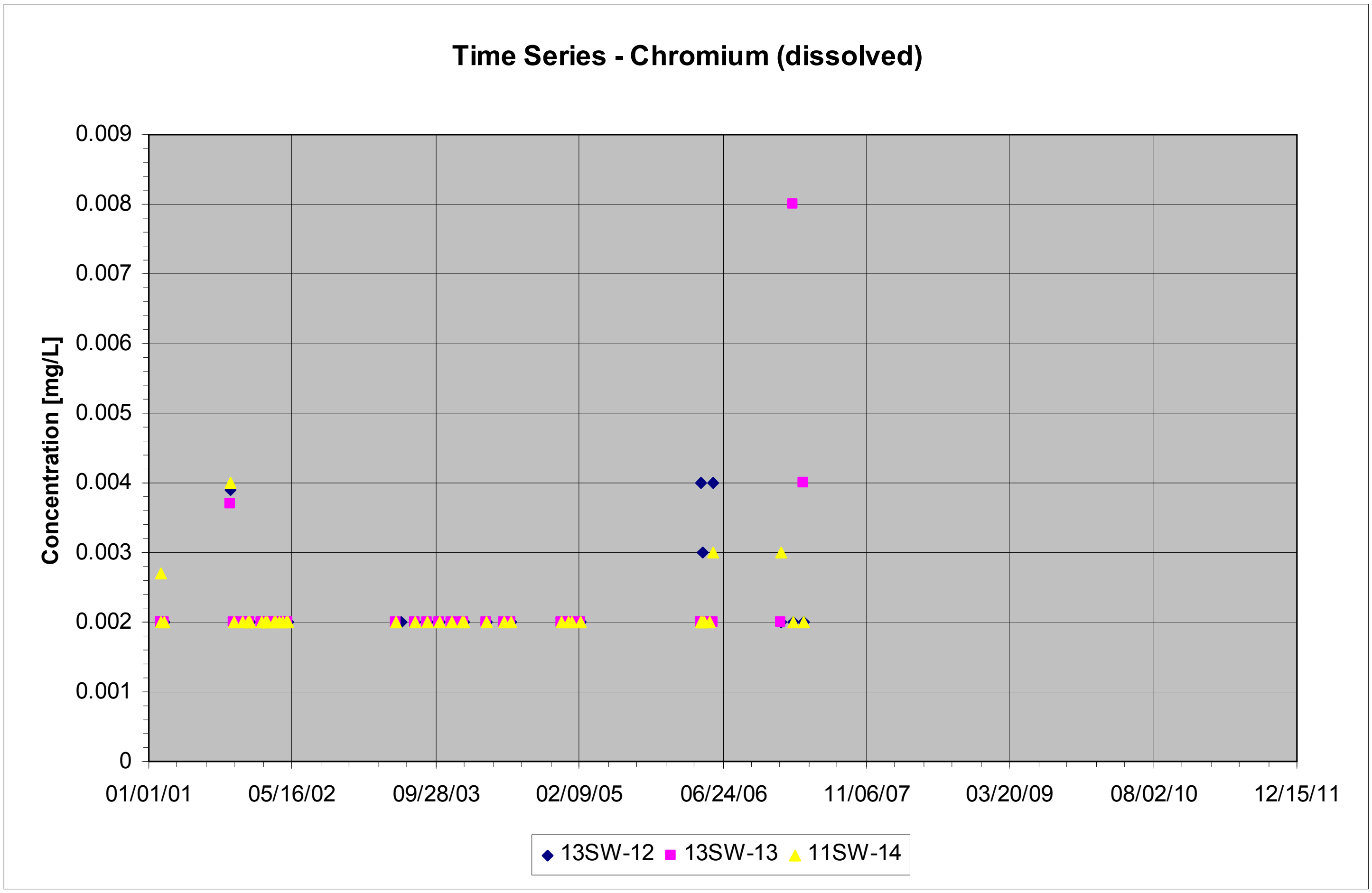

Time Series - Boron (total)
Concentration [mg/L]
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

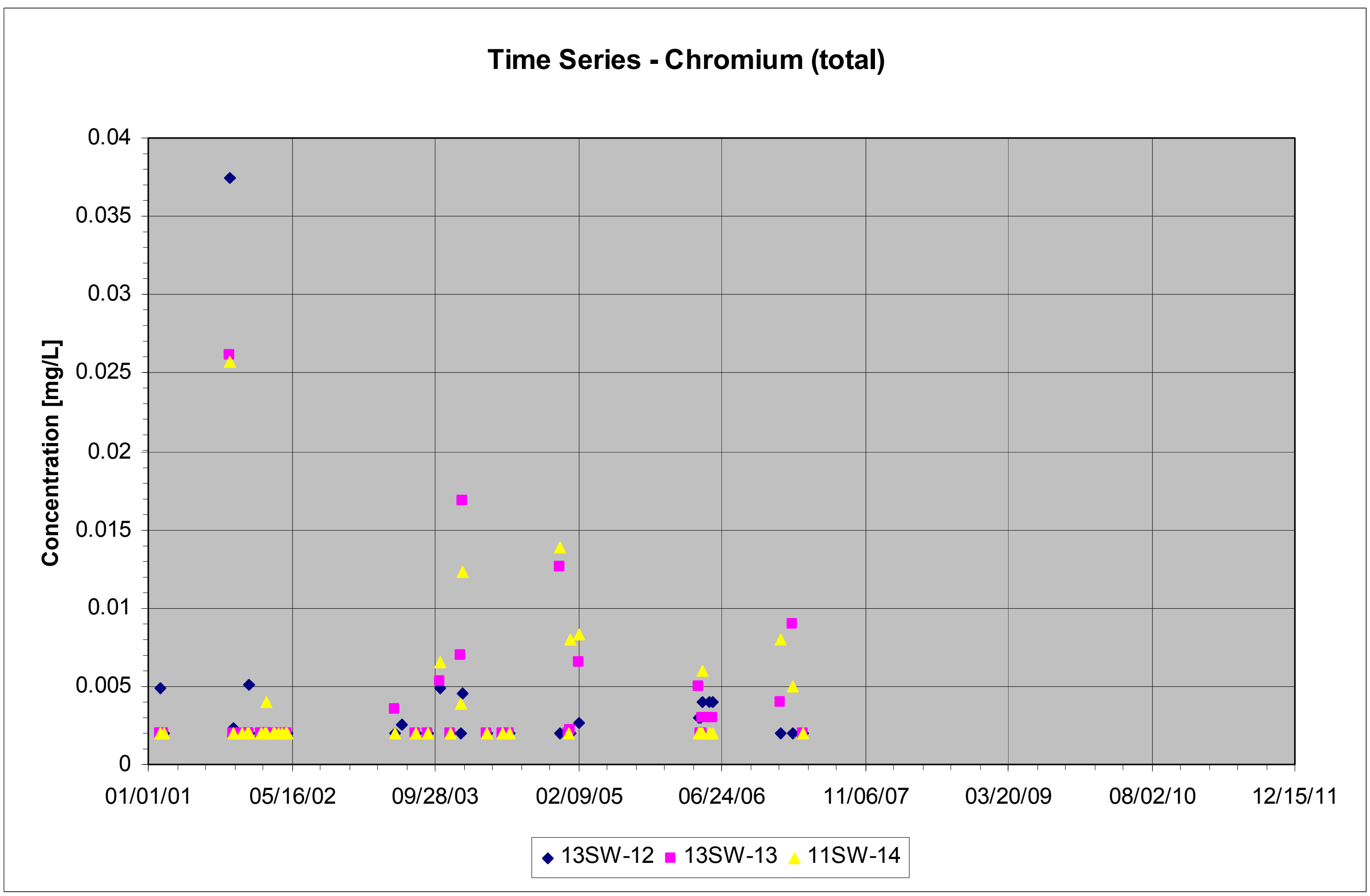


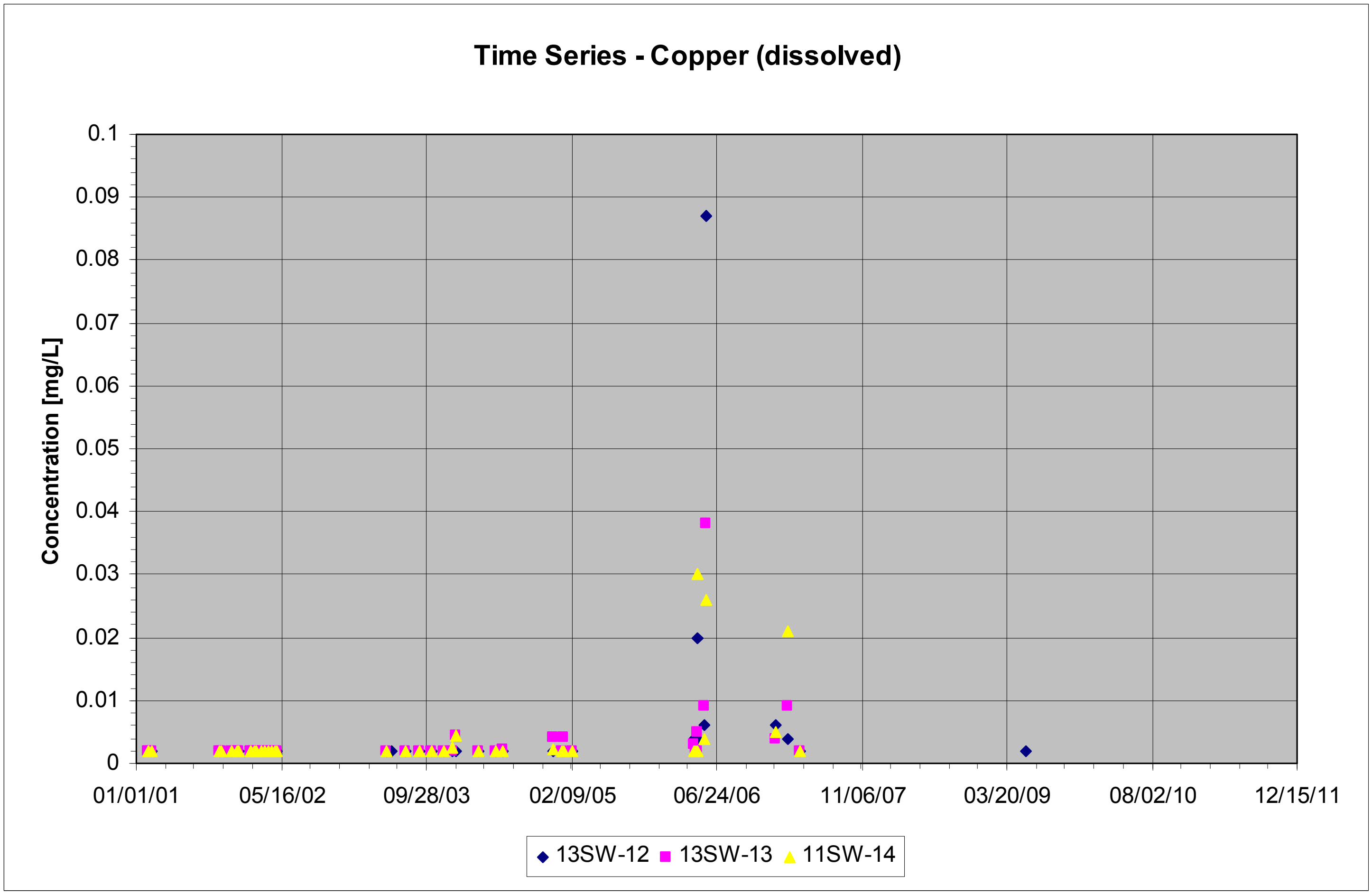
Time Series - Cadmium (dissolved)
Concentration [mg/L]
0.0025
0.002
0.0015
0.001
0.0005
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



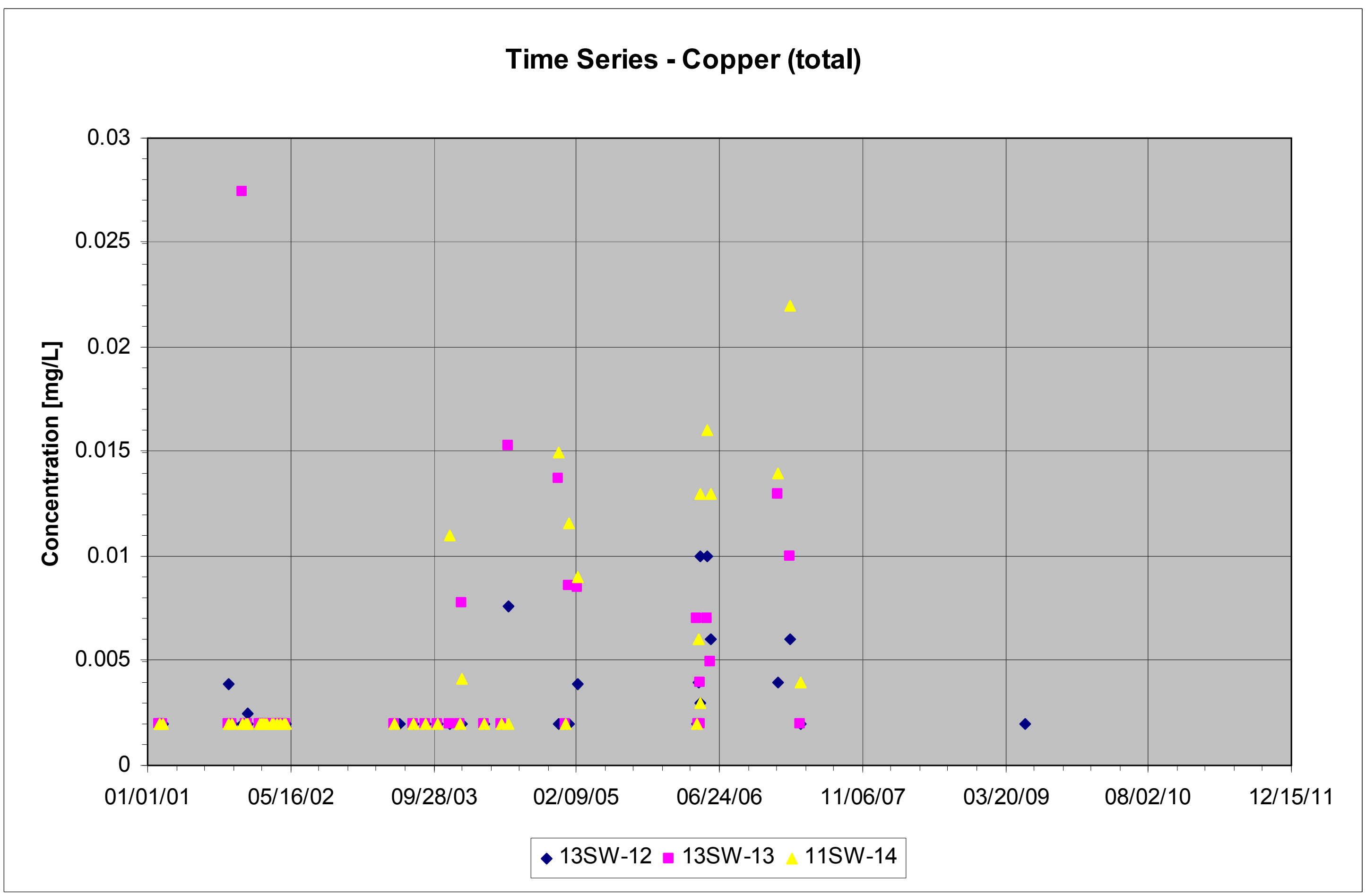
Time Series - Cadmium (total)
Concentration [mg/L]
0.0025
0.002
0.0015
0.001
0.0005
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14


Time Series - Chromium (dissolved)
Concentration [mg/L]
0.009
0.008
0.007
0.006
0.005
0.004
0.003
0.002
0.001
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

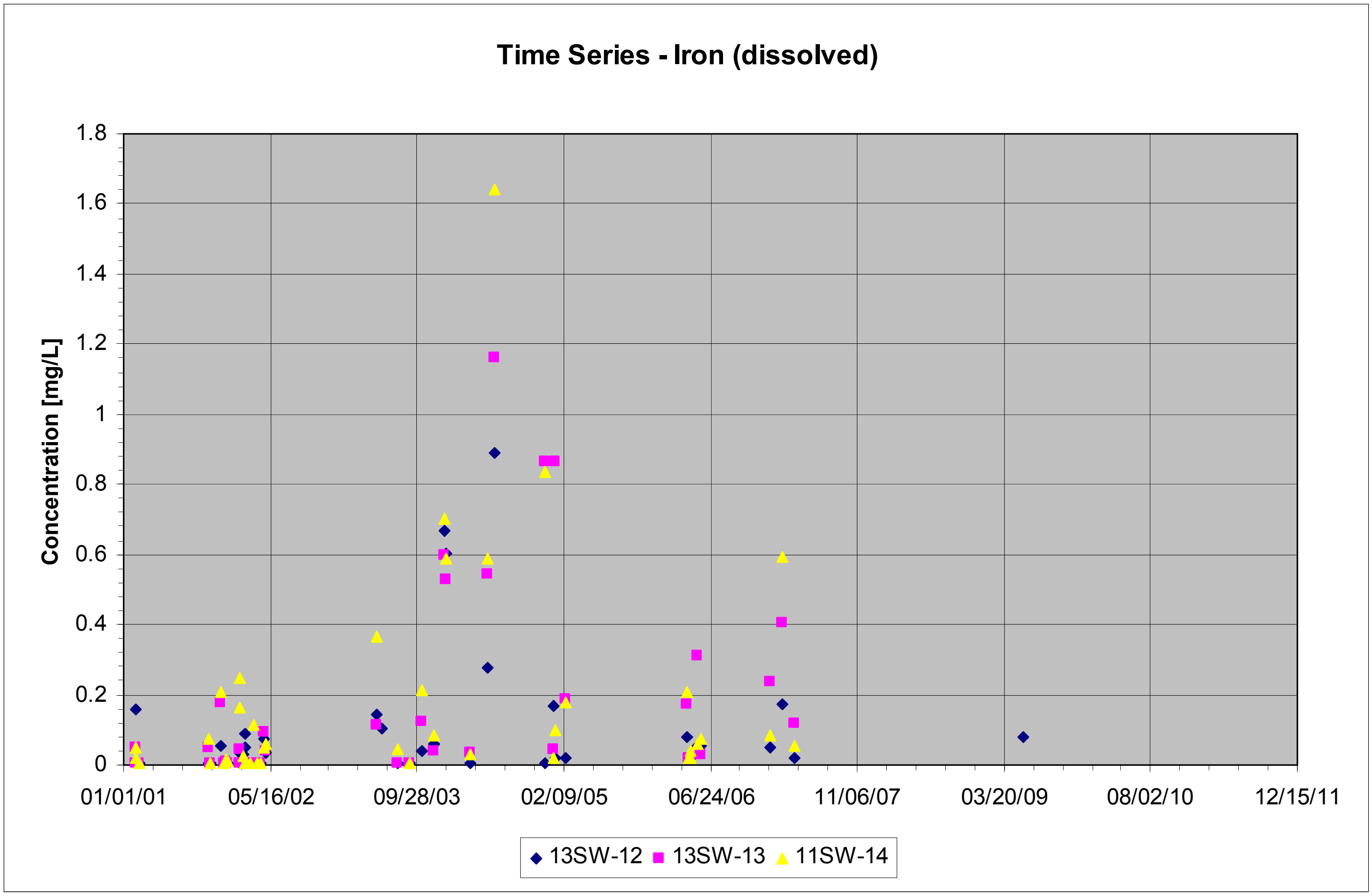

Time Series - Chromium (total)
Concentration [mg/L]
0.04
0.035
0.03
0.025
0.02
0.015
0.01
0.005
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

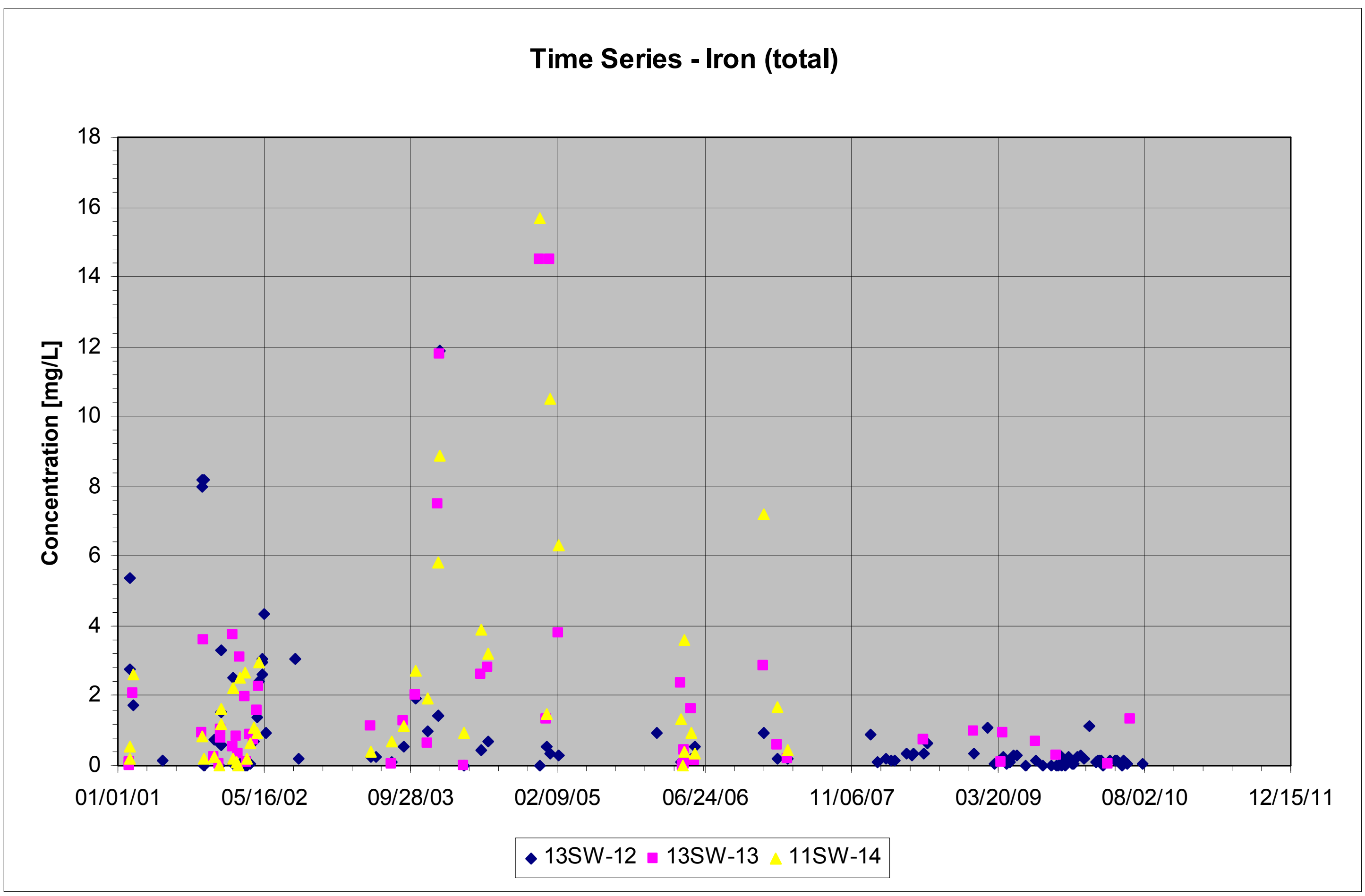


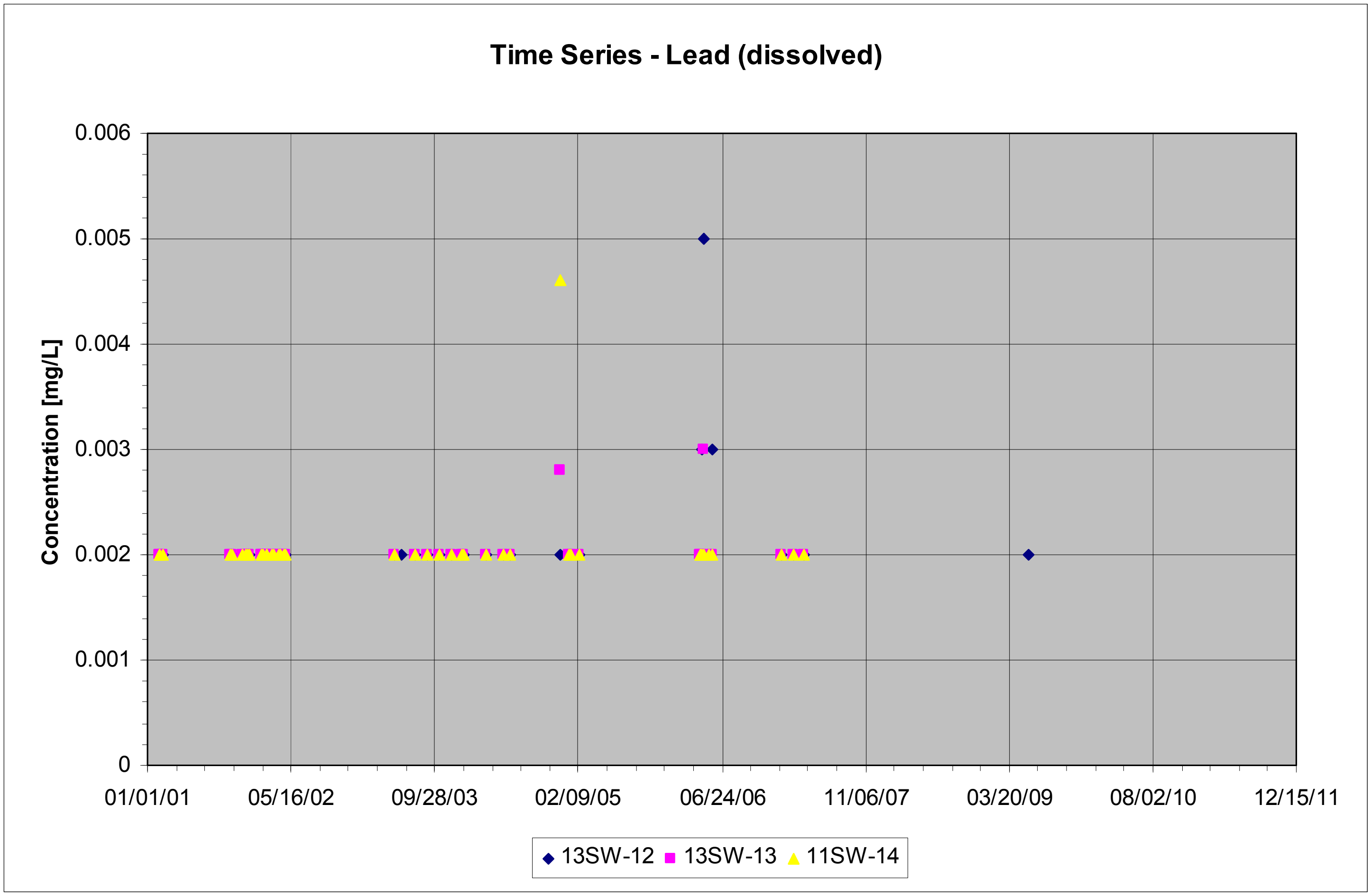
Time Series - Copper (dissolved)
Concentration [mg/L]
0.1
0.09
0.08
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



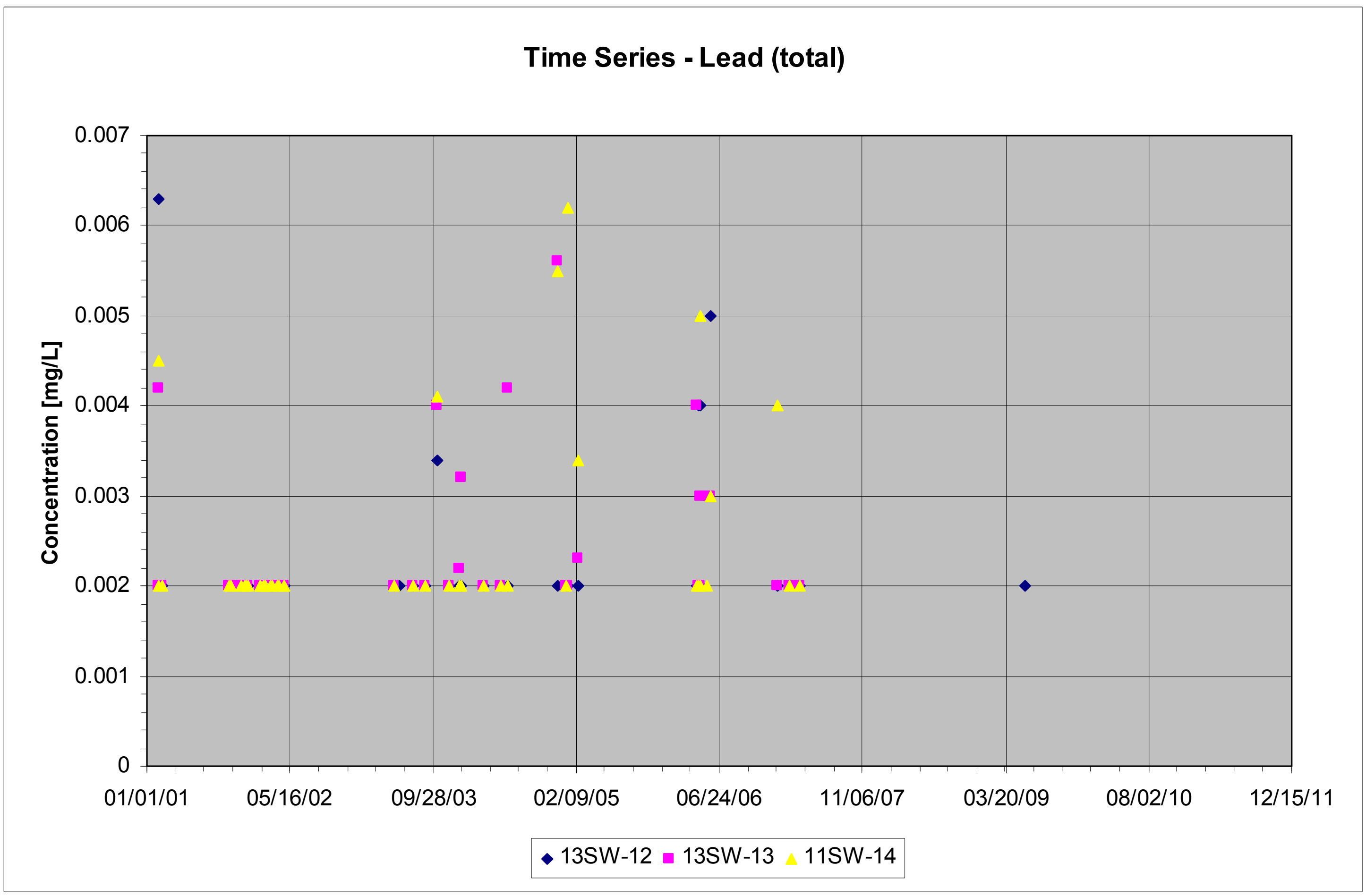
Time Series - Copper (total)
Concentration [mg/L]
0.03
0.025
0.02
0.015
0.01
0.005
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14


Time Series - Iron (dissolved)
Concentration [mg/L]
1.8
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

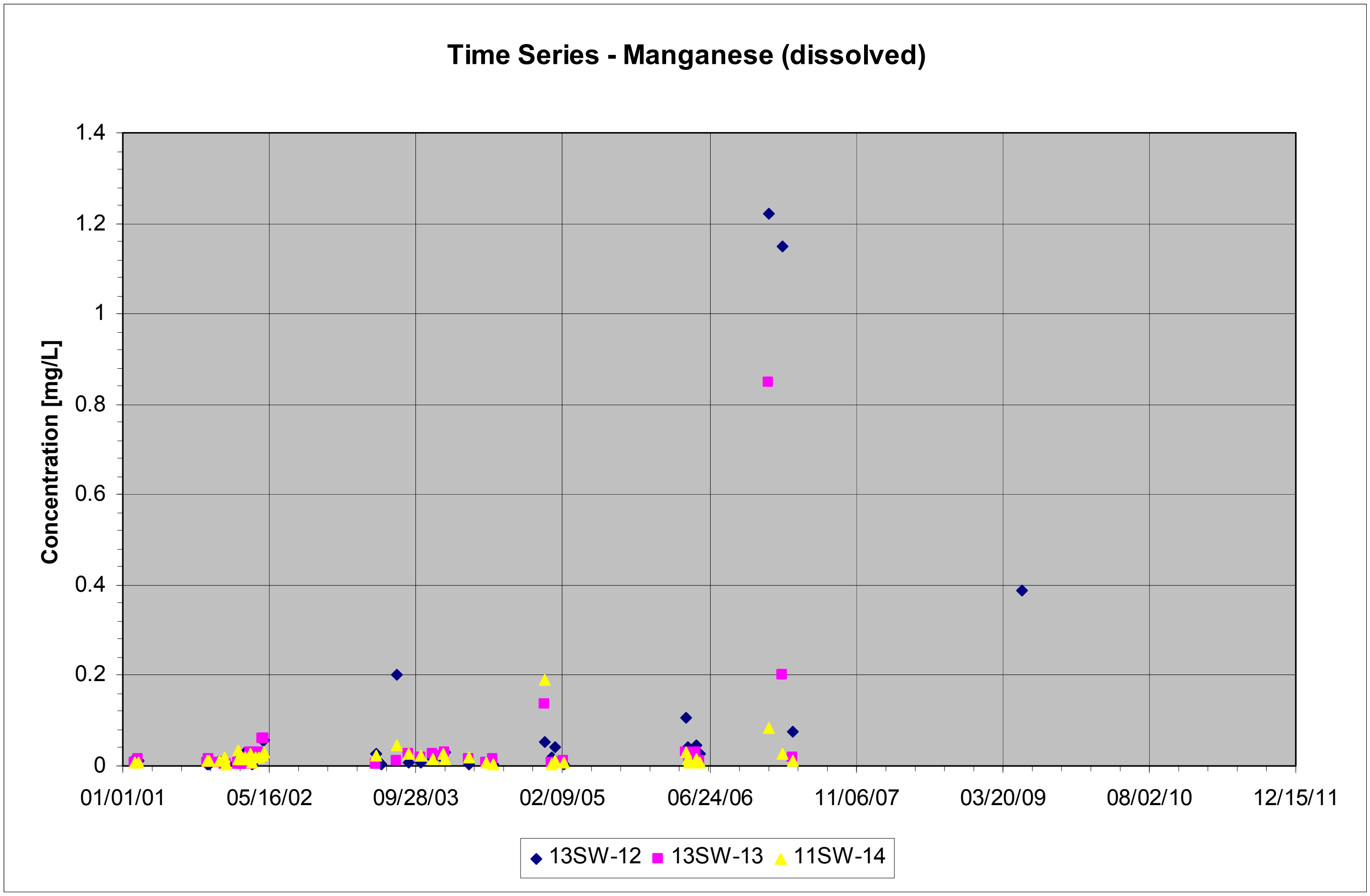

Time Series - Iron (total)
Concentration [mg/L]
18
16
14
12
10
8
6
4
2
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



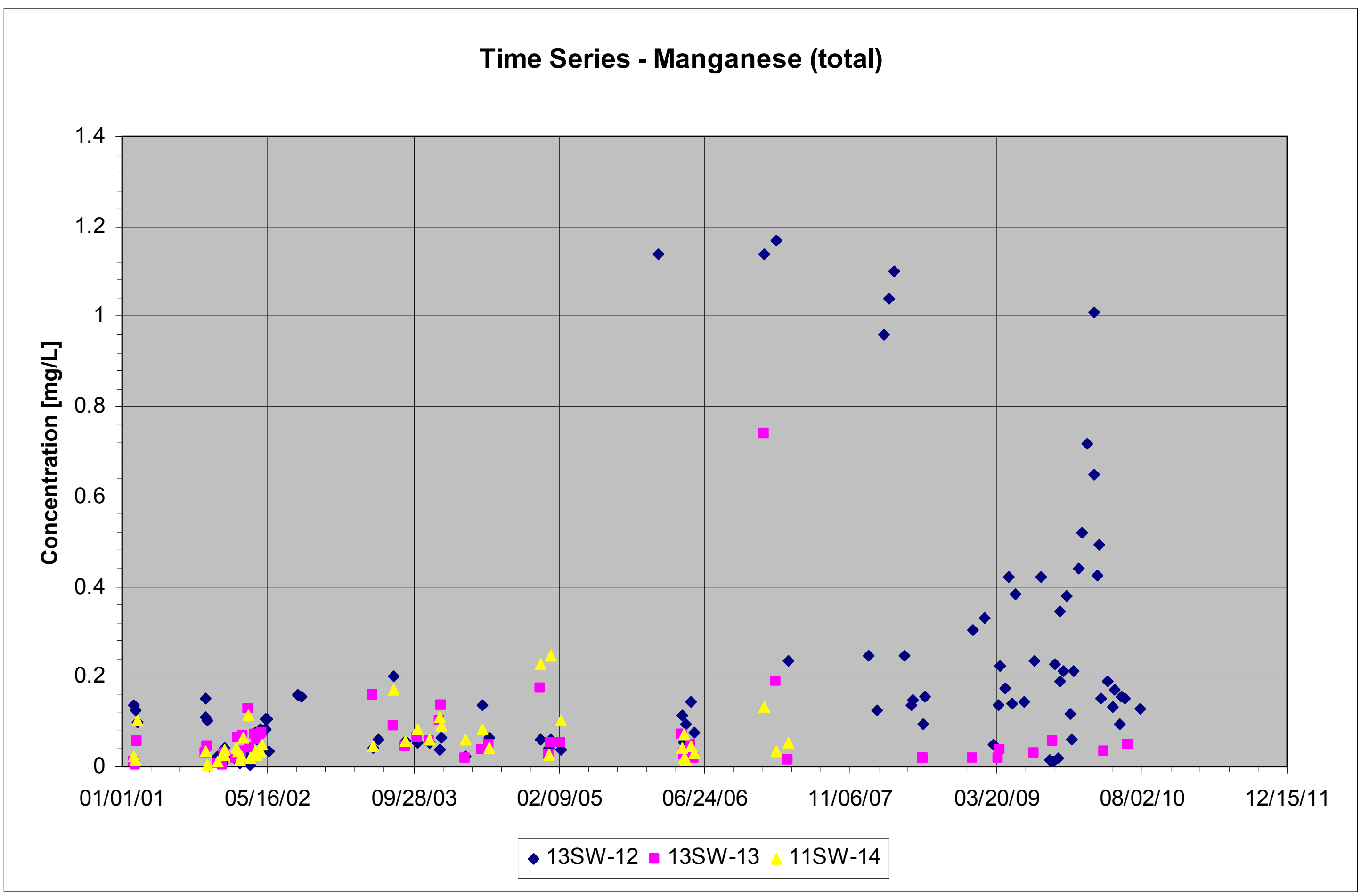
Time Series - Lead (dissolved)
0.006
0.005
0.004
0.003
0.002
0.001
0
Concentration [mg/L]
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



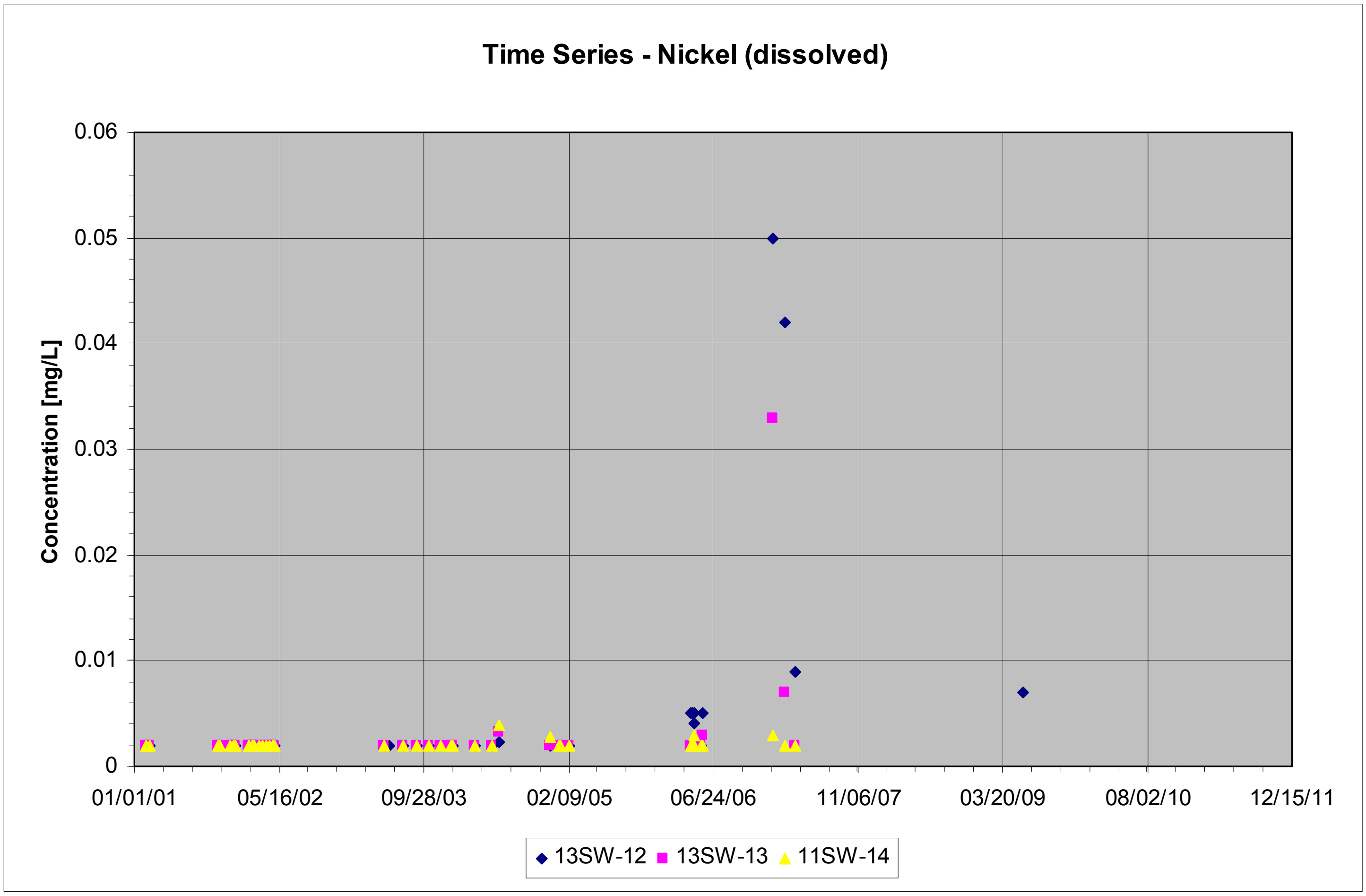
Time Series - Lead (total)
0.007
0.006
0.005
0.004
0.003
0.002
0.001
0
Concentration [mg/L]
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



Time Series - Manganese (dissolved)
Concentration [mg/L]
1.4
1.2
1
0.8
0.6
0.4
0.2
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



Time Series - Manganese (total)
Concentration [mg/L]
1.4
1.2
1
0.8
0.6
0.4
0.2
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



Time Series - Nickel (dissolved)
Concentration [mg/L]
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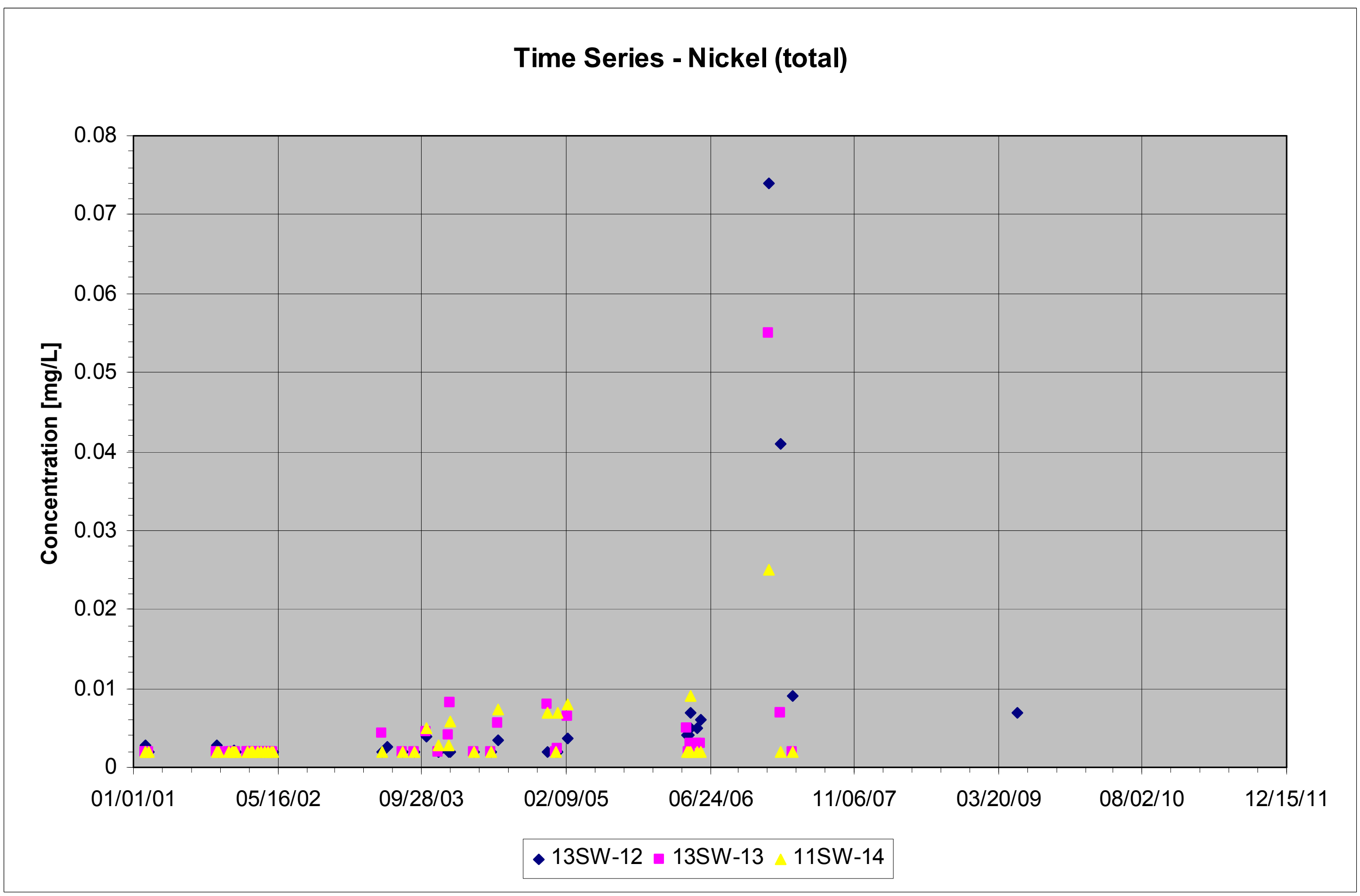
0.01
0.02
0.03
0.04
0.05
0.06
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14



Time Series - Nickel (total)
Concentration [mg/L]
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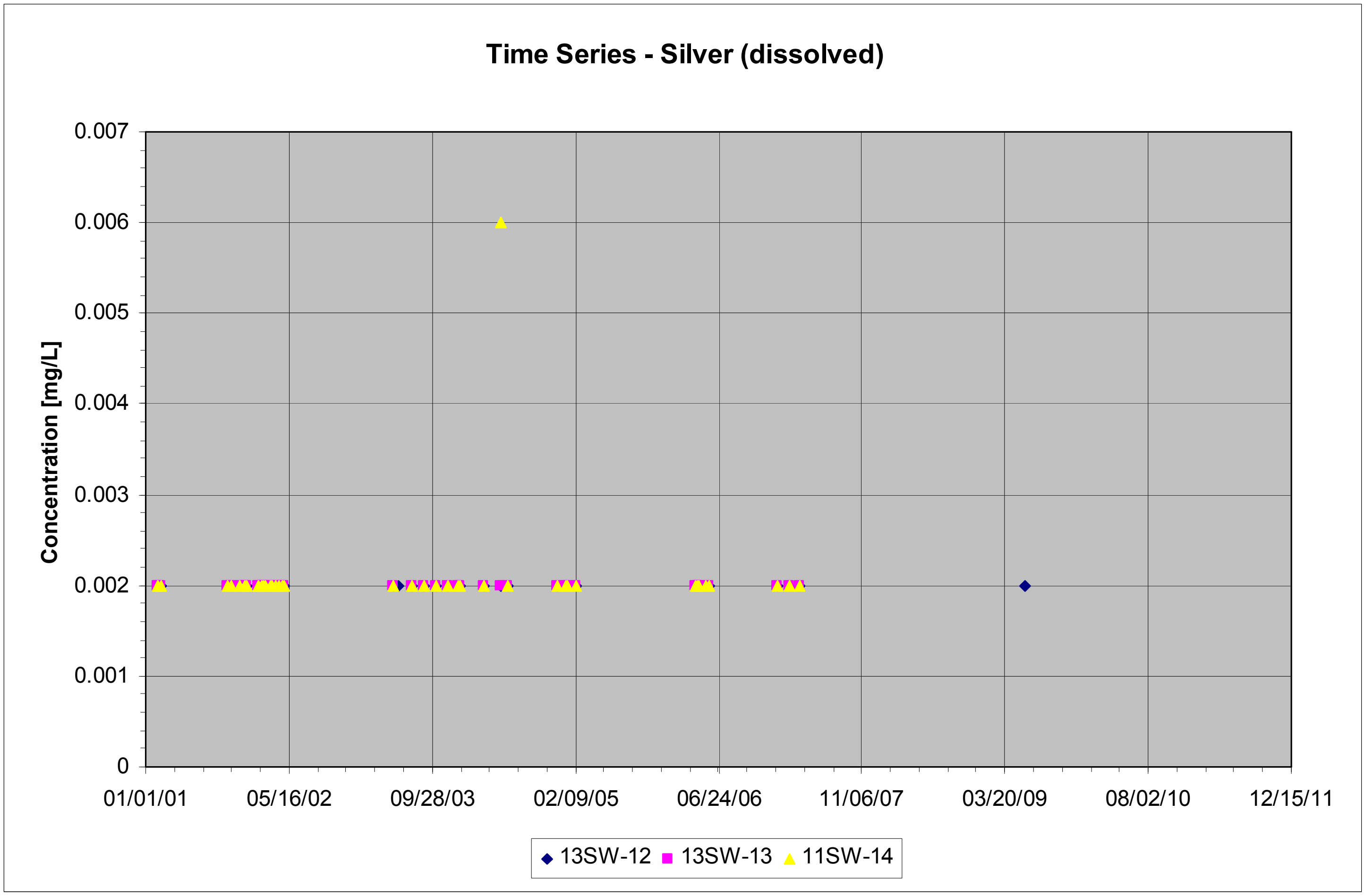
0.01
0.02
0.03
0.04
0.05
0.06
0.07
0.08
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14


Time Series - Silver (dissolved)
Concentration [mg/L]
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0.006
0.005
0.004
0.003
0.002
0.001
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
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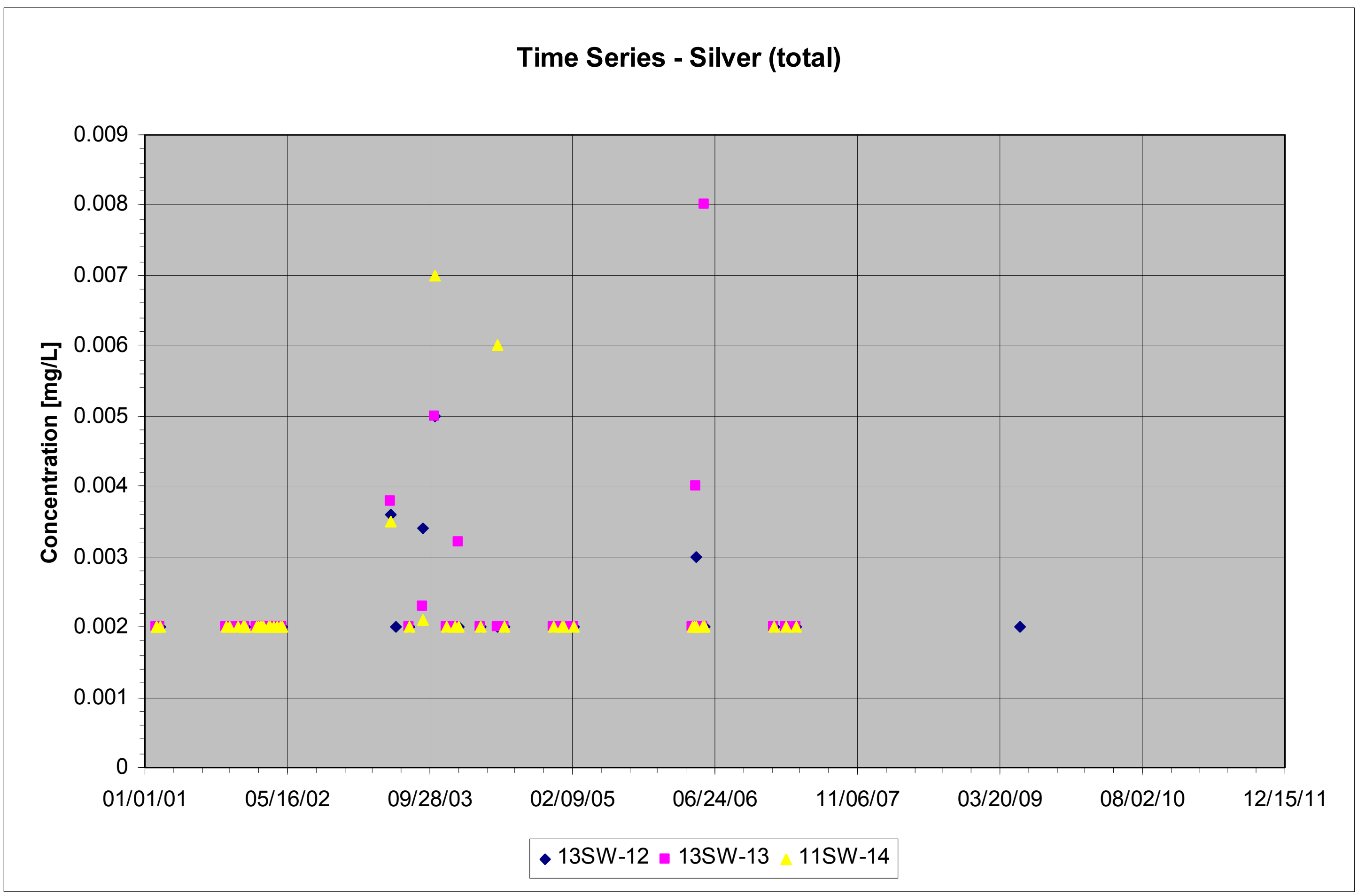
13SW-13
11SW-14


Time Series - Silver (total)
Concentration [mg/L]
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0.008
0.007
0.006
0.005
0.004
0.003
0.002
0.001
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
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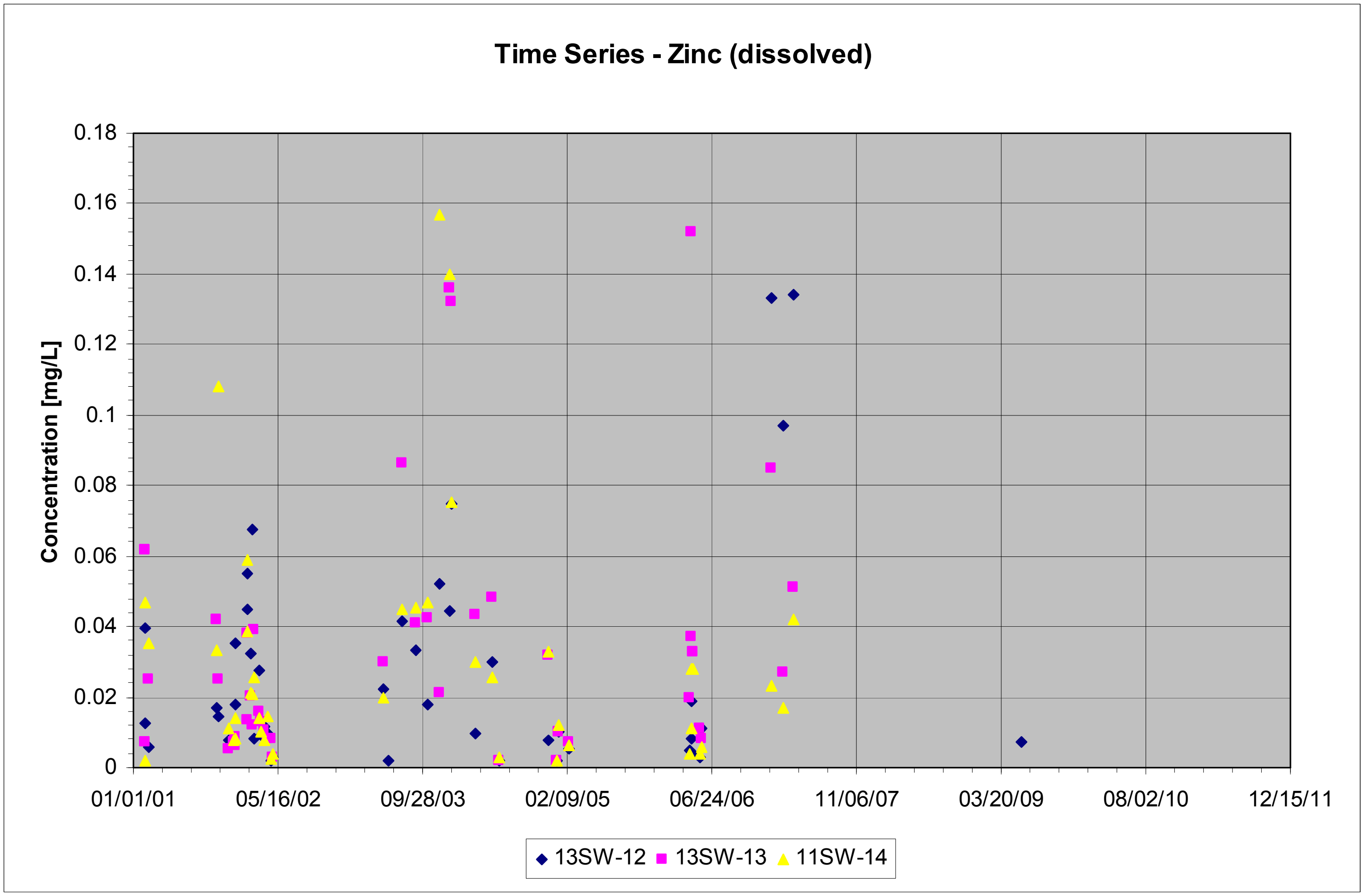
13SW-13
11SW-14


Time Series - Zinc (dissolved)
Concentration [mg/L]
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0.16
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
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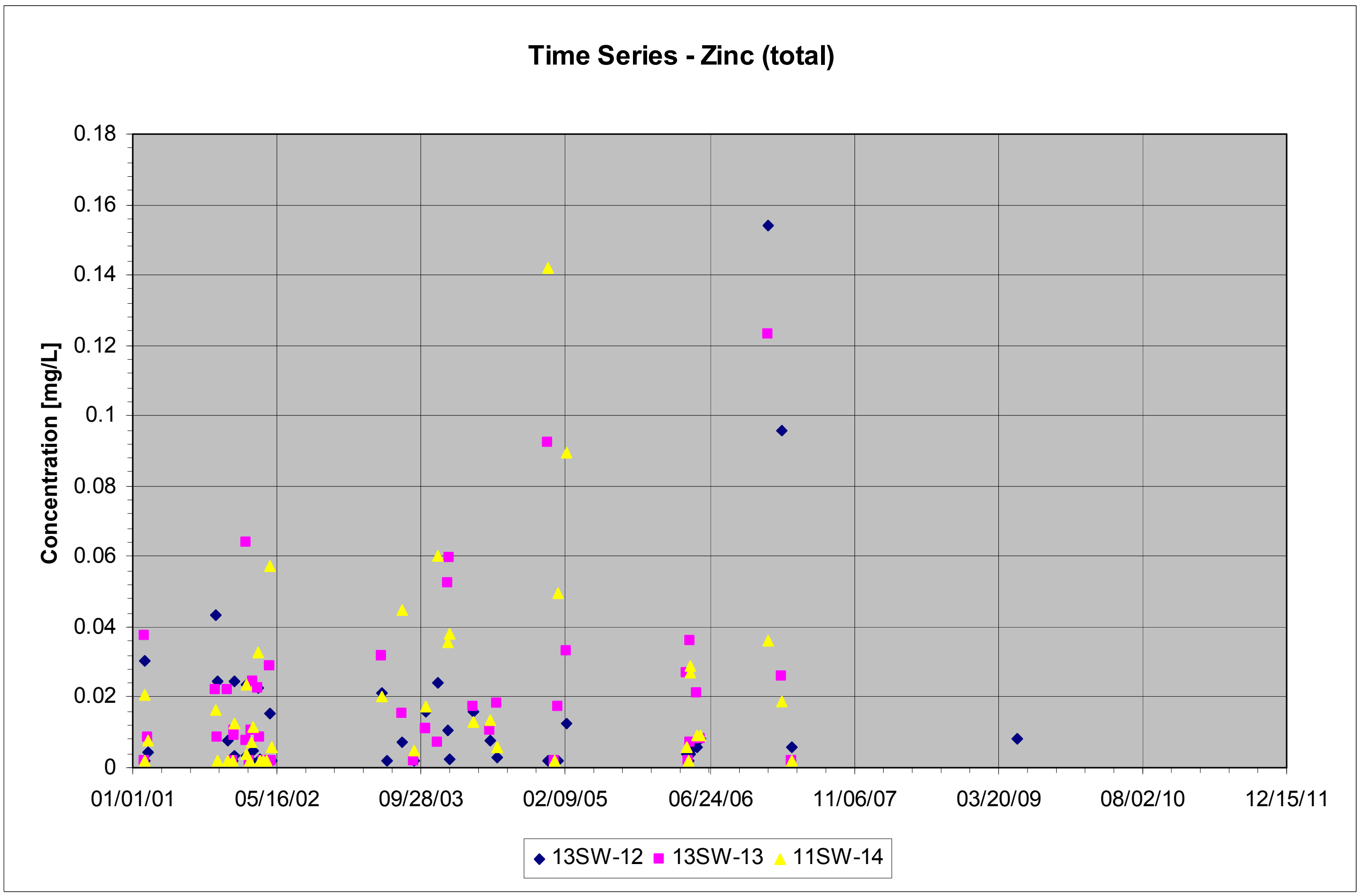
13SW-13
11SW-14


Time Series - Zinc (total)
Concentration [mg/L]
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0.16
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
01/01/01
05/16/02
09/28/03
02/09/05
06/24/06
11/06/07
03/20/09
08/02/10
12/15/11
13SW-12
13SW-13
11SW-14

APPENDIX B – EXHIBIT 4

Report for Fish and Macroinvertebrate Sampling for Bioassessment Monitoring of West Fork Busseron Creek

Prepared for:

Peabody Energy
Evansville, Indiana

Prepared by:

ENVIRON International Corporation
Denver, Colorado

Date:
September 10, 2010

Project No:
2024989A

Contents

Executive Summary

Peabody Midwest Mining, LLC (Peabody) has reconstructed a portion of the West Fork Busseron Creek, near Farmersburg, IN, (Sullivan County) in response to mitigation of mining activities for Farmersburg Mine. ENVIRON International Corporation (ENVIRON) conducted a biological stream survey that incorporated fish, benthos, and habitat evaluation specific for the Farmersburg Mine and West Fork Busseron Creek Mitigation (WFBCM) area with a comparison to an upstream reference site located within an undisturbed reach of WFBC. This monitoring event served as an interim status check on stream biota to document recovery and in-stream biological development following stream reconstruction.

Water quality field measurements and selected water chemistry results indicated a slight decrease downstream in concentration of conductivity and all major ions except potassium within the WFBCM. Dissolved oxygen, pH, and temperature showed typical diurnal fluctuation common the exposed stream systems. Habitat evaluations based on the Qualitative Habitat Evaluation Index (QHEI) and USEPA Rapid Bioassessment Protocols (USEPA 1989, 1999) resulted in habitat assessment scores that indicated mid-suboptimal habitat conditions for both the reference and the WFBCM.

A total of 15 different fish species were identified in the WFBCM. Fish survey results indicated a minnow-based assemblage at the reference area compared to a sunfish-based assemblage in the upper portion of the reconstructed reach, and a sunfish and minnow-based assemblage in the lower portion of the reconstructed reach. The fish community was dominated by insectivores and only the largemouth bass represented a top carnivore/predator species at the reference site and the WFBCM. Fish Index of Biotic Integrity (IBI) scores ranged from 42-44 indicating fair biotic status at the reference site and ranged from 40-44 for the WFBCM indicating negligible difference in the fish assemblage between the reference and WFBCM.

The benthic macroinvertebrate survey was conducted at the reference site and both WFBCM sites using the multi-habitat approach with riffle samples being kept separate from vegetation/debris dam samples. A total of 89 different taxonomic entries were identified, which represented specimens from the major aquatic insect groups plus a presence of clams, snails, worms, and crustaceans. Organisms representing the Diptera-Chironomidae (flies and midges) dominated the macroinvertebrate collections at all sites. Macroinvertebrate IBI results based on USEPA (1989), for use with a reference collection, indicated slightly lower biological integrity conditions at both sites within the WFBCM for the riffle samples, and only at the upstream portion of the WFBCM for the vegetation/debris dam samples (IBI score less than 79% of the reference score). The downstream vegetation/debris dam sample was over 100% of the reference IBI score indicating no loss of biological integrity or condition.

Associations between attributes of habitat features, the fish community, and the macroinvertebrate community within the WFBCM indicate typical hydraulic function and biological functions of a healthy stream system are present. A continuation of the functional aspects of the hydrologic pattern in combination with maturity of the channel, bank, and riparian area of the WFBCM will form the basis and future development of fish, benthos, and other aquatic-based communities. Based on the findings of this study, it is believed that over time, the compositional

structure of the fish and benthic macroinvertebrate assemblages will mimic reference conditions and a minnow based community can develop within the WFBCM.

1. Background & Objectives

This monitoring project is to serve as a status check for recruitment and establishment of biota for the reconstructed portion of West Fork Busseron Creek known as the West Fork Busseron Creek Mitigation (WFBCM) area. The WFBCM area is approximately 7,825 feet in length and was constructed in response to mitigation of mining activities for Farmersburg Mine. The stream reconstruction plans incorporated current aspects and understanding of hydrology and stream morphology to enhance the ecological benefits of the stream specific to the gradient and geographical area. ENVIRON conducted a biological stream survey June 29-July 1, 2010 to provide biological information as a temporal benchmark to demonstrate the gradual succession within the mitigation area towards pre-mining conditions.

2. Methods

2.1 General

The stream survey of the WFBCM was based on selected physico-chemical constituents, habitat attributes, and resident biological community parameters for benthic macroinvertebrates, and fish. Survey methods were based on Rankin (1989), IDEM (2006) and U.S. Environmental Protection Agency (USEPA 1989, 1999) for the Qualitative Habitat Evaluation Index (QHEI), and habitat bioassessment, respectively; USEPA (1989, 1999) and Ohio Environmental Protection Agency (OEPA) for benthic macroinvertebrates and fish collection and evaluation.

Field work was conducted by general progression from downstream to upstream, implementing tasks sequentially based on technical considerations. For example, water samples and *in situ* water quality analyses were conducted prior to all field activities so as not to alter water quality due to in-stream activity, fish collections were conducted prior to other activities so as not to disturb fish in preferred habitats, and habitat assessments were conducted after all in-stream activities to best familiarize team members with habitat conditions.

2.2 Sample Locations

ENVIRON personnel toured the reconstruction site on June 29, 2010 to determine most appropriate locations for macroinvertebrate and fish collection. One upstream reference site (WFBCU1) located outside the WFBCM and two downstream sites (WFBCR2 and WFBCR3) within the WFBCM were selected for benthic macroinvertebrate collection (Figure 1). Sites WFBCR2 and WFBCR3 were also used for fish collection with reference conditions represented by fish survey data from immediately below the WFBCM and conducted prior to stream relocation and construction (Three Rivers Environmental 2003).

Site location, corresponding latitude and longitude, was determined with a hand-held GPS. All samples collected were recorded in bound field logbooks to facilitate sample tracking. Labeled water chemistry samples were shipped the same day as collected to one of several Test America analytical laboratories depending upon the suite of analytes to be evaluated. Preserved benthic macroinvertebrate samples were stored with internal and external labels and shipped from the study site to EcoAnalysts, Inc (Moscow, ID) for taxonomic analysis. Sample collection quality assurance and quality control (QA/QC) objectives were met as no samples were lost and all results can be traced back to the correct spatial location of collection.

2.3 Physical/Chemical Parameters

2.3.1 Habitat Quality

Habitat assessments were conducted in the upstream reach, WFBCU1, and downstream segments of WFBCR2 and WFBCR 3 on June 29, 2010. Habitat quality was assessed for the entire 150 meter (m) study reach and was documented using the visual based approach presented in *Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers* USEPA 1999. The Indiana Qualitative Habitat Evaluation Index (QHEI) was also determined as a composite of the entire study reach and was based on Qualitative Habitat Evaluation Index (QHEI) Standard Operating Procedure (Document S-001-OWQ-A-BS-06-S-R1 Dec 2006).

2.3.2 Water Quality and Flow

A Horiba Model U-10 multi-probe meter was used for *in situ* water quality at all locations where biological samples and water chemistry samples are collected. Daily calibrations consistent with manufacturers' recommendations were conducted prior to use and following use at the end of the day to verify proper operation and maintain consistency in meter readings.

The following *in situ* parameters were assessed:

- Dissolved Oxygen (mg/L)
- pH (s.u.)
- Conductivity (µmhos/cm)
- Temperature (°C).

Instantaneous discharge was determined by the incremental flow method at the center of each study reach with the aid of a standard top-setting rod and Marsh-McBirney Model 2000 flow meter. A minimum of 10 increments were measured for depth and water velocity across a transect perpendicular to the stream flow, and then combined to determine total instantaneous discharge.

2.4 Biological Survey

2.4.1 Fish

ENVIRON has reviewed a pre-mining fish census report (Three Rivers Environmental 2003) and duplicated the fish survey efforts as much as possible in order to maximize comparison of results. ENVIRON surveyed sites WFBCR2 and WFBCR3 within the WFBCM with battery powered backpack electroshock fishing unit using standard and accepted protocols as follows:

1. One fish survey location was no closer than 30 meters of the downstream terminus of the WFBCM, and the other fish survey location was no closer than 100 ft from the upstream end of the WFBCM.

2. Fish sampling was conducted at each of the two WFBCM sites in a stream reach that was 150 meters in length, which was a minimum of 15 times the wetted width of the stream.
3. An electroshock sampling time of a minimum 40 minutes was the target sampling effort at each site to best match the fishing effort reported by Three Rivers Environmental (2003) at the reference site
4. Standard fish shocking methods were followed to meet data quality objectives of comparable data to previous survey efforts. Block nets were set at the lower and upper ends of the measured reach; shocking proceeded in an upstream direction and all pool, riffle, run, and backwaters were sampled. All attempts were made to maintain captured fish alive in temperature appropriate site water within coolers and holding tanks for analysis. The entire study reach was sampled by electroshocking twice; and all fish were returned to WFBCM area unharmed following specimen analysis and data recording.

2.4.2 Fish Data Collection

The following information was documented on in-house fish survey field forms or field logbook:

1. Site information to include West Fork Busseron Creek, Sullivan County, Indiana, date and time at study reach, and personnel on-site.
2. Sample site information to include GPS coordinates of downstream and upstream ends of stream survey reach, stream length of survey reach, and general stream morphology (average depth, velocity, and instantaneous discharge).
3. Water quality information include pH (s.u.) dissolved oxygen (mg/L), specific conductivity (µmhos/cm), and temperature (°C) upon arrival at the site and at end of day.
4. Water chemistry information to include laboratory alkalinity, laboratory conductivity, total dissolved solids, and the major ionic composition of the water to include calcium (Ca), magnesium (Mg), sodium (Na), potassium (K), bicarbonate (HCO_3), chloride (Cl) and sulfate (SO_4).
5. Fish information to include species identification, enumeration, and length measurements. Fish weights will be taken for at least five fish from each of a representative size class per species (total size classes per species and weights dependent upon capture and number of fish) to facilitate estimation of biomass. All fish captured were identified and enumerated.
6. All fish were inspected for anomalies, deformities, or indications of disease and any such observations were recorded.

2.4.2 Benthic Macroinvertebrates

Benthic macroinvertebrates were collected from within the WFBCM area at locations within the fish survey reach sections, and from an undisturbed reach in West Fork Busseron Creek upstream of the WFBCM. The upstream sample of benthic macroinvertebrates provided an in-stream reference of the benthic community composition and structure for the WFBCM reach. A qualitative multi-habitat sampling scheme was followed that is consistent with several state and federally accepted and approved macroinvertebrate bioassessment sampling methods. Benthic macroinvertebrate sampling within the WFBCM and undisturbed upstream West Fork Busseron Creek site included the following:

1. Benthic macroinvertebrate sampling consisted of kick net and dip net sample collections from representative locations of each of the key habitats in each sample reach. Riffle habitat samples were collected from a 1 square meter area using either a kick net with 500 *u*m mesh for or a D-frame kick net with 500 µm mesh. The D-frame kick net with 500 µm mesh was used for vegetation sweeps and debris dam habitat type sampling. A 3 square meter area of riffle and an equal area estimated for the streamside vegetation/debris dam habitat were sampled in each study reach. The collection of streamside vegetation and woody debris samples was timed to approximate and equal the riffle sampling effort. Vegetation and debris sampling included the collection and shaking of individual debris clumps and dams or sweep samples of material for a minimum of 2 minutes before removing residual material and transferring the remaining material and organisms to labeled sample containers. The number of dip net samples collected from the streamside vegetation and woody debris habitat type were recorded in a bound field logbook.
2. Samples from within the same habitat type were combined as a composite in one quart plastic bottles, and field-preserved with 95 percent ethanol. Thus, there was a riffle sample container, and a vegetation/debris dam sample container for each study reach. All samples were identified by habitat type, sample station and date collected, and contained both internal and external labels.
3. Locational data such as GPS coordinates of the habitat collections, reach name, sample identification, date and time were recorded in the bound field logbook. Photographic records of representative habitat sample types were included.
4. Samples were shipped overnight to EcoAnalysts (Moscow, ID) for taxonomic analysis and metric calculation. Organisms were identified to the species level whenever possible. Benthic community metrics common to bioassessment indices and characterization of the benthic community were calculated.

3. Results

3.1 Physical/Chemical Parameters

3.1.1 Habitat Quality

Habitat assessment scores using the visual based USEPA (1999) habitat assessment score sheets for low gradient streams resulted in a range of habitat scores from 123 to 135 for sites WFBCR3 (reconstructed area) and WFBCU1 (upstream reference area), respectively. Composite QHEI scores, calculated following guidelines of IDEM (2006), for the WFBCM study reaches were 50 for WFBCR3 downstream; 52 for WFBCR2 in the upper portion of the reconstructed zone. The QHEI score was 53 for the WFBCU1 reference area, upstream of the reconstructed zone (Table 1). Habitat metrics values for each evaluation are presented in Attachment 1.

Habitat scores for the USEPA (1999) protocol indicate mid-suboptimal conditions at all three sites. It is important to note that while the USEPA forms were primarily designed to assess habitat quality of natural streams, they were used here because of the Rosgen stream design approach applied during the planning stage of the stream reconstruction. One goal of the Rosgen stream design approach is to re-create a high quality natural hydrologic stream condition. Thus, a successful reconstructed stream should have high values for several metrics that assess conditions of channel sinuosity, flow status, natural stream patterns, and riparian vegetative protection and width. Low metric values within the USEPA habitat assessment were typically assigned to those metrics associated with temporal aspects of stream hydraulics such as epifaunal substrate and cover, substrate characterization, and sediment transport/deposition. Because the WFBCM is relatively young (less than 3 years) it is anticipated that following several years of further hydraulic development and stabilization within the stream channel coincident with continued vegetative development of the riparian zone, the habitat will continue to improve towards optimal conditions.

The QHEI results were consistent with the USEPA habitat assessment with respect to little difference in QHEI score between the reference WFBCU1 area and the two reconstructed study reaches of WFBCM. The average QHEI score of 17 transects in WFBC study during 2003 (prior to reconstruction) was 54.7 (Three Rivers Environmental 2003). The Three Rivers Environmental (2003) sites were located downstream of the bridge over WFBC at County Road 950N. The QHEI scores of 50 and 52 attained for the present study following reconstruction demonstrate habitat conditions are comparable to those initially present prior to reconstruction. In addition, the QHEI scores within the WFBCM agree well and are comparable to the QHEI score of 53 for the upstream WFBCU1 reference area.

3.1.2 Water Quality and Flow

Flow measurements and water quality determinations for dissolved oxygen, temperature, conductivity, and pH measured *in situ* at the sample locations are shown in Table 2. Flow was approximately 4.5 cubic feet per second (cfs) higher in the WFBCM than the 5.25 cfs measured upstream at the WFBCU1 site. Temperature showed a typical pattern of warming during the day at all sites and was generally warmer in the WFBCM than upstream at the WFFCU1 site. Dissolved oxygen concentrations at all sites indicated high oxygen availability to aquatic organisms, and pH ranged from 7.7 s.u., upstream to 9.0 s.u., at the downstream end of the WFBCM. Both dissolved oxygen and pH showed a range typical of a response to diurnal fluctuations in water temperature.

Analytical results of water chemistry samples collected at all three sampling sites are shown in Table 3. Concentrations of the selected constituents are within the range expected for the stream and site. The data show a slight decline in all constituents downstream, with the highest concentrations determined at the upstream WFBCU1 reference area.

3.2 Biological Survey

3.2.1 Fish

A total of 15 different fish species were identified from the electroshock survey of the WFBCM on June 30 – July 1, 2010. Twelve species were found in the upstream reach at WFBCR2 and 13 species were found at WFBCR3. During this survey, fish species found only at WFBCR1 included steelcolor shiner (3 specimens) and white sucker (10 specimens). Fish species found only in the downstream WFBCR3 reach included two quillback specimens, silverjaw minnow (31 organisms), and a single spotted sunfish. A summary of the fish survey including number and total biomass for each species identified for the WFBCR2 and WFBCR3 study reaches of the reconstructed stream area is shown in Table 4, along with fish survey results conducted near WFBCU1 in 2002 prior to stream reconstruction (Three Rivers Environmental 2003). A listing of individual fish specimens captured along with weights and length data is included in Attachment 2.

The assemblage of fish represented species common to Indiana and frequently encountered in small headwater to moderate sized streams (Simon and Dufour 1997). Sunfish (bluegill, green sunfish, longear sunfish) were the dominant group represented at both the WFBCR2 and WFBCR3 followed by the largemouth bass as a single species and members of the minnow family as a group. The fish assemblage was dominated by insectivore species (10 of the 15 total species encountered) with the largemouth bass representing the only carnivore/piscivore recorded. Evaluation of the feeding strategies for the additional fish species show on Table 4 as reported by Three Rivers Environmental (2003) and not encountered in this study show the same pattern. All additional fish identified by Three Rivers Environmental (2003) were insectivores, except the Mississippi silvery minnow (omnivore), resulting in the largemouth bass being the only carnivore/piscivore encountered in this portion of WFBC prior to and following reconstruction.

Bioassessment results based on the Index of Biotic Integrity (IBI) for fish captured in the reconstructed zone at WFBCR2 and WFBCR3 are shown at the bottom of Table 4 as well as the IBI score. The individual metric values for the IBI based on the fish assemblage at WFBCR2, WFBCR3, and the 2002 samples reported by Three Rivers Environmental (2003) are shown in Table 5. IBI metric values and final IBI scores for WFBCR2 and WFBCR3 were based on protocols in Simon and Dufour (1997) for the Eastern Corn Belt Plain Region in Indiana which includes the Sullivan County area. IBI scores were 44 for WFBCR2 and 40 for WFBCR3 and are nearly identical to scores of 44 (upstream) and 42 (downstream) reported by Three Rivers Environmental (2003) for samples collected prior to stream reconstruction. A comparison of the IBI scores indicates the WFBCM has presently attained the level of biotic integrity that existed in WFBC prior to stream relocation and construction. Based on Simon and Dufour (1997) IBI scores ranging from 40-44 are rated as Fair, with attributes that include loss of intolerant species, decrease in species number, a highly skewed trophic structure, and the older age classes of top predator may be rare. The assemblage of fish collected at both WFBCR2 and WFBCR3 show these attributes by:

1. A general lack of fish species considered sensitive or intolerant for the Eastern Corn Belt Plain,
2. A skewed trophic structure by a dominance of insectivore species, few omnivores, and a single carnivore,
3. The low total biomass of largemouth bass, the single top predator combined with only two specimens out of 53 attaining adult lengths of 25 and 27 cm, and
4. Low end of the predicted number of species for the region (although sufficient for the maximum metric value).

Similar results were indicated by the Three Rivers Environmental data for 2002 where no largemouth bass were recorded from the "upstream" of County Road 950N and only one largemouth bass specimen, attaining a biomass of 2.9 grams, was reported at the "downstream" site. Based on a length:weight relationship of the largemouth bass specimens captured at WFBCR2 and WFBCR3, a 2.9 gram largemouth bass would be 6.4 cm long and likely not an adult.

3.2.2 Benthic Macroinvertebrates

Benthic macroinvertebrate samples were collected on June 29, 2010 using the multi-habitat sampling approach that resulted in a sample from fast, medium, and slow riffle areas in one container, and vegetation sweeps and debris dam samples in a second container from each of the WFBCU1 (reference), WFBCR2 and WFBCR3 study reaches. A taxonomic listing with enumeration data for each riffle and multi-habitat sample collected from the study reaches is presented in Attachment 2 along with a listing of general community structure and composition metrics for macroinvertebrate samples.

The benthic organisms identified in the collections from WFBC included taxa for all major aquatic insect groups plus mussels and clams (Bivalvia), snails (Gastropods), worms (Annelida) and crustaceans. Specimens represented taxa common to the region and no rare, endangered, or otherwise special status species were encountered. The overall listing the benthic macroinvertebrate organisms identified from WFBC indicates the greatest number of taxa representing the Diptera (flies and midges) especially the chironomids, the Coleoptera (aquatic beetles) and Gastropods (snails). These types of organisms are generally considered tolerant of physical stress and occur in a wide range of water quality conditions. Organisms that are typically considered sensitive to degradation of water quality and unstable or poor habitat conditions were not very diverse, were poorly represented, or absent from the collected samples. For example, these more sensitive organisms would include the EPT taxa consisting of members of Ephemeroptera (three species recorded), Trichoptera (four genera reported) and

Plecoptera (absent). However, the natural low gradient, warm ambient temperatures, and generally sandy and small particle substrate material characteristic of the WFBC watershed would generally favor an assemblage of the more tolerant taxa.

Evaluation of macroinvertebrate data from WFBC was performed with USEPA Protocol III (USEPA 1989), which uses a multimetric index and scoring system to compare a reference benthic assemblage to the benthic assemblage from one or more study sites to determine biotic integrity or impairment status. In this case, the reference assemblage is represented by the WFBCU1 macroinvertebrate samples. The WFBCU1 site is within a forested area of the watershed that has generally been undisturbed and natural for the past 50 years or more (personal communication, Richard Williams Peabody Energy, June 29, 2010). This site is a more appropriate site-specific reference condition for assessing the biotic integrity of the WFBCR2 and WFBCR3 than elsewhere within the same or alternative watershed.

Results of the USEPA (1989) bioassessment method for macroinvertebrates are presented in Table 6 for the composite riffle samples and in Table 7 for the multi-habitat (vegetation sweep and debris dams) composite sample from each study site. Final multi-metric scores for the riffle samples indicated the biotic integrity of the benthic assemblage at WFBCR2 was less than the samples collected at WFBCU1 (reference). The biotic integrity at WFBCR3 was equal to the reference reach, WFBCU1 for the riffle habitat (Table 6). However, final multi-metric scores for the vegetation/debris dam samples indicated the biotic integrity of the benthic assemblage at both reaches within the reconstructed area was less than the biotic integrity indicated by samples from the reference site (Table 7). The biotic index approach is not always sensitive to subtle shifts in taxonomic composition due to habitat differences other physic-chemical attributes. For example, the survey data show the most abundant organism for riffle samples was the caddisfly, *Cheumatopsyche* (Trichoptera), at the reference area; the fly *Pseudochironomus* (Diptera-Chironomidae) at WFBCR2, and the aquatic beetle *Berosus* (Coleoptera) at WFBCR3. The value for the Percent Contribution of the Dominant Taxon metric focuses only the magnitude of the relative abundance data and does not consider the ecological difference between the caddisfly (sensitive) and the Diptera (tolerant).

Results of the macroinvertebrate survey demonstrate that factors, such as invertebrate drift and primary and secondary productivity within the WFBCM, support complete life cycles and redistribution and colonization of aquatic insects. In addition, the rate of development implied by the relative level of biotic integrity determined at WFBCR2 and WFBCR3 is consistent with generally accepted expectations of aquatic insect community recovery within 5-7 years following episodic catastrophic events such as dam failures and floods. Further development of the benthic community can be expected. However, development of the benthic community (and fishery) will be more dependent upon the hydrologic patterns that continue to redistribute movable sediment material to form stable habitats and the progressive maturity of the bank and riparian features of the reconstructed portion of WFBC.

4. Summary

This study focused on the structural aspects of the major biological components of a stream system to demonstrate the successful relocation and construction of a stream reach of WFBC. Key features of fish and benthic macroinvertebrate community structure and composition were subjected to bioassessment techniques using multiple community metrics. The community metrics incorporate autecological information and form a basis for ecological interpretation with respect to health and biological integrity of the stream. Implications from the structural aspects of the biotic communities can also provide insight to various functional aspects of a stream and this association can further demonstrate successful relocation and construction of the WFBCM reach.

Evaluation of the stream morphology and riparian features that support the biological communities of a stream were found to be comparable using the QHEI (IDEM 2006) and USEPA habitat assessment methods. Interpretation of the habitat bioassessment scores indicated the physical condition of all study sites was suboptimal (Table 1). Individual metric values implied a lack of riffle habitat, uniform substrate composition within the riffles, and a general lack of substrate diversity prevented optimal conditions. These same attributes, in addition to a poorly developed bank and riparian buffer zone, prevented optimal conditions within the WFBCM (Attachment 1). Because of the undisturbed nature of the reference area (WFBCU1), a significant change to optimal habitat conditions is unlikely. However, within the reconstructed reach (sites WFBCR2 and WFBCR3) the design features and reclamation efforts applied to the stream and riparian area are in the process of maturing by means of the seasonal hydrologic patterns within the stream channel (hydraulic distribution of sediment and armoring of hard substrates) and growth of seeded and planted vegetation along the banks and riparian area. No differences in water quality or water chemistry between the reference area and the reconstructed area were indentified that would strongly influence the physical habitat (Table 2 and Table 3). Based on the current status, future habitat evaluations are likely to trend towards optimal conditions in the reconstructed portion of WFBC.

Key findings from the biological evaluation using bioassessment techniques for the fish and macroinvertebrate survey data in the WFBCM include the following:

1. Bioassessment results for fish indicate comparable biotic index values of 40 and 44 for the WFBCR3 and WFBCR2 sites, respectively, which overlap the biotic index values of 42 and 44 for the WFBCU1 reference area (Table 5). The biotic index value indicates negligible difference in biotic integrity of the fish assemblage between the reference site and reconstructed stream sites. Common features of the fishery between the reference and reconstructed area include the presence of only the largemouth bass as the single species representing a top carnivore, with nearly all other species being strict insectivores. This represents a skewed trophic structure and is consistent with characteristics of biotic integrity scores in the range of 42-44. Key differences between the fish assemblage at the reference site and reconstructed study sites that are not reflected by the metrics of the biotic index involve distribution and abundance among the fish species encountered. For example, Table 4 shows the fish community at WFBCR2 could be characterized as a sunfish dominated (numerically and biomass contribution) assemblage consisting of bluegill, green sunfish, and longear sunfish; compared to WFBCR3

that would be characterized as a sunfish (bluegill and longear as biomass) and minnow (silverjaw minnow, suckermouth minnow numerically) dominated assemblage; compared to the reference site WFBCU1 that would be considered a minnow dominated fish assemblage (bluntnose minnow, creek chub, and silverjaw minnow). Since sunfish, and especially green sunfish, are known to be good colonizers of new and available habitats, it is likely that as the channel and riparian corridor of the WFBCM develops and matures into a channel with stable riffle substrates, a shift towards a minnow dominated community can be expected.

2. The benthic macroinvertebrate survey indicated colonization, recruitment, and the development of a benthic community has occurred in the WFBCM following stream reconstruction. Taxa present included the major aquatic insect groups in addition to other organisms such as clams, snails, and crustaceans for a total of 89 different taxonomic entities. The benthic community at all sites exhibited a high number of taxa representing flies and midges (Diptera and Chironomids) and aquatic beetles (Coleoptera). However, differences among the reference sites and study sites were present as demonstrated by the shift in taxa of the most dominant organism (see Attachment 2) and indicated by the Community Loss metric for the riffle samples (Table 6). The Community Loss metric value represents the decimal percent of taxa that are not common between the reference WFBCU1 site and the study sites, indicating only 30% of the taxa at WFBCR2 were also found at the reference site while WFBCR3 has approximately 63% of taxa common with WFBCU1. Bioassessment results for the benthic macroinvertebrates indicated the biotic integrity in the WFBCM was not as high as indicated by the benthic community at the WFBCU1 reference area. Other community structure metrics, and evaluation of habitat scores suggest that the lower biotic integrity at sites WFBCR2 and WFBCR3 may be associated with the physical habitat (unstable, and underdeveloped substrates), and the progression of community development. For example, the Shannon Diversity value for the benthic macroinvertebrates assemblages from the riffle habitat is 3.11 at WFBCR2 and 4.04 at WFBCR3 compared to 2.91 at the reference site. Biological diversity is typically higher during a colonization and development period when habitat features are unstable and changing allowing a number of different organisms to be present, compared to later when habitat features are stable and the community structure dynamics have limited the number of organisms to those that are adapted to the existing conditions. The higher diversity values observed in the WFBCM imply the physical nature of the riffles are changing and have yet to stabilize. The diversity of the benthic macroinvertebrates for the vegetation/debris dam samples is similarly related to the available habitat. In this instance, the vegetation/debris dam habitat is relatively absent in the reconstructed area (open channel, few obstructions) as compared to WFBCU1. (closed channel, many obstructions) as shown by photographs in Attachment 3. Differences in diversity in this case are associated with a lack of complexity in the habitat. It is anticipated that the biotic integrity of the benthic macroinvertebrate community will increase in the WFBCM as the stream channel, substrates, and bank/riparian corridor become for mature and stable.

3. The expected increase in biotic integrity for both the fish and benthic macroinvertebrate communities demonstrates an expected response to one of the

functional aspects of a stream system. An important hydraulic function of a stream includes sediment transport and substrate development, especially following episodic disturbances when the channel is new. The current status of the fish and benthic macroinvertebrate communities demonstrate the temporal aspects of seasonal hydrologic patterns in progressively forming more stable substrate and channel features has been occurring since construction. As mentioned above, the continued function of sediment transport and substrate development with the WFBCM is the basis for the progression towards stable riffle habitats and the shift from a sunfish dominated fish community to a minnow dominated community as observed prior to construction.

4. Other key features of streams include biological functions such as energy transfer and carbon processing between biological communities, productivity and respiration rates, decomposition, and nutrient cycling, which work in concert with hydraulic functions. Measurement of these functional aspects was not a target of this investigation. However, features of the biological data that were collected implicate these stream functions occur. The presence of more than one type of biological community representing two major trophic levels (macroinvertebrates as secondary consumers and fish as tertiary consumers) in the absence of a sustained fish stocking program is evidence that biological functions exist and are active in WFBCM. More specifically, a review of the functional metrics for the riffle sample benthic macroinvertebrates (Appendix 2) indicates the assemblage at each of the three sample sites include organisms from all feeding strategies in proportions that provide insight to sources of primary production and carbon processing. For example, differences in the percent abundance of filterers and gatherers among the sites suggest food resources are primarily suspended, small particles of debris that are being transported within the water column (high percentage of filterers) while in the reconstructed reaches the available food resources also exist in or on the surface of the substrates (high percentage of gatherers). A review of the percent contribution of scrapers and shredders suggest that algal growth is moderate in the reference area (low contribution of scrapers), nearly absent at WFBCR2, and likely abundant at WFBCR3. The indications regarding algal growth (primary producers) suggested by the distribution of functional feeding groups corresponds with the habitat metric scores and features at the study sites. The reference site exhibits mature vegetation along the bank and riparian area that provides shade which can limit the development of permanent algae growth in the riffles (low to moderate scraper contribution), the movable substrates and unstable nature of the substrates at WFBCR2 that would severely reduce both the growth and access to algal growth (very low scraper contribution), and the more stable riffle substrate combined with and lack of mature vegetation to provide shade allows greater algal growth at WFBCR3 (high scraper contribution). A review of the percent contribution of scrapers from the multihabitat samples also corresponds with the physical features of the study sites and provides insight regarding the location and sources of primary production by algae growth. The high contribution of scrapers in debris dams at the reference site corresponds with the greater availability of this type of habitat due to inputs of leaves, sticks and debris from the bank and riparian area. The contribution of scrapers at WFBCR2 from the vegetation sweep/debris dam samples is much

greater than observed in the riffles at this site due to the extensive cattail growth within the channel at this site (algae growth attached to the submerged portion of the cattails), while at WFBCR3 no such extensive cattail habitat was present (lowest scraper contribution). Another example demonstrates the biological function addressing transfer of energy/biomass from one trophic level to another. The WFBCM sites studied did not exhibit extensive algae growth at any site implying high inputs or excess nutrient availability does not occur on a sustained basis. This preludes the abundance of certain fish species (central stoneroller, largescale stoneroller, and southern redbelly dace) that are common to small streams in this geographical region (Simone and Dufour 1989). The predominant feeding strategy for the fish assemblage encountered at all WFBC study sites was the category insectivore, which represented 89% of the fish captured at WFBCR2 and 73% of the fish captured at WFBCR3. A balanced biological function of energy/carbon transfer between trophic level is implied by differences in the estimated density of fish at WFBCR2 and WVBCR3 that showed the same pattern for difference in the estimated density of benthic macroinvertebrates from these sites. Site WFBCR2 exhibited higher estimated densities for both fish and macroinvertebrate than were estimated for site WFBCR3. At site WFBCR2 the estimated fish density and combined habitat macroinvertebrate density was 1.05 fish per square meter (fish/M^2) and 2,250 insects/ M^2 compared to 0.79 fish/M^2 and 1,709 insects/ M^2 at WFBCR3. A more appropriate functional evaluation of energy/biomass transfer would be conducted with biomass, but those data were not available. However, what may be simply coincidental given the many factors involved regarding fish size and age, preferences in available diet, and macroinvertebrate life stage, it is interesting to note that the magnitude of change in density from WFBCR2 compared to WFBCR3 for fish (24.7% lower) and for insects (24.0% lower) were nearly identical.

5. References

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Table 1. West Fork Busseron Creek Habitat Survey Summary

Sample Site	Date	QHEI Score[1]	EPA Score[2]	EPA Score Description
WFBCU1	29-Jun-10	53	135	Mid-Suboptimal
WFBCR2	29-Jun-10	52	127	Mid-Suboptimal
WFBCR3	29-Jun-10	50	123	Mid-Suboptimal

Notes:
1. QHEI scores as per IDEM Draft Qualitative Habitat Evaluation Index Protocol 2006.
2. EPA score as per Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers USEPA 1999

Table 2. West Fork Busseron Creek *In Situ* Field Measurements

Sample Site	Date	Time	Latitude (N)	Longitude (W)	pH (su)	Conductivity (uS)	Dissolved Oxygen (mg/L)	Temperature (˚C)	Instream flow (cfs)
WFBCU1	29-Jun-10	1130	39.2473	87.3644	7.76	690	7.7	27.0	-
	29-Jun-10	1320	39.2477	87.3643	-	-	-	-	5.25
	1-Jul-10	1655			8.42	685	8.5	29.1	-
WFBCR2	29-Jun-10	1415	39.2364	87.3614	8.52	474	8.9	28.7	-
	29-Jun-10	1525			-	-	-	-	9.95
	1-Jul-10	930			8.37	513	8.7	25.3	-
	1-Jul-10	1610			9.15	507	12.2	29.4	-
WFBCR3	29-Jun-10	1720	39.2316	87.3593	8.52	476	8.4	29.7	-
	29-Jun-10	1820			-	-	-	-	9.94
	30-Jun-10	950			8.07	486	8.8	25.3	-
	1-Jul-10	1628			9.06	470	9.9	31.6	-

Notes:
1. Instream flow calculated from instream velocity/depth measurements

Table 3. West Fork Busseron Creek Water Chemistry Analytical Results

Sample Site	Date	Chloride (mg/L)	Sulfate (mg/L)	TDS (mg/L)	Alkalinity (as $CaCO_3$ mg/L)	Calcium (mg/L)	Magnesium (mg/L)	Potassium (mg/L)	Sodium (mg/L)	Hardness[1] (mg/L)	Site-Specific SO_4 Criteria[2] (mg/L)
WFBCU1	1-Jul-10	15.5	217	435	120	57.9	28.2	3.63	35.0	259	1,485
WFBCR2	1-Jul-10	13.0	147	312	97.2	41.1	20.4	3.38	26.1	186	1,119
WFBCR3	1-Jul-10	11.9	136	288	96.3	40.5	19.3	3.45	22.6	180	1,058

Notes:
1. Hardness is calculated from magnesium and calcium concentrations.
2. Calculated using hardness and chloride values according to 37 IAC 2-1-6.

Table 4. West Fork Busseron Creek Fish Survey Results

Common Name	*Genus Species*	ENVIRON				Three Rivers Environmental[1]			
		Site: WFBCR2 W Fk Busseron Cr - Summer 2010		Site: WFBCR3 W Fk Busseron Cr - Summer 2010		Site: "Upstream" W Fk Busseron Cr - Summer 2002		Site: "Downstream" W Fk Busseron Cr - Summer 2002	
		Count	Total Biomass (g)	Count	Total Biomass (g)	Count	Total Biomass (g)	Count	Total Biomass (g)
Blackstripe topminnow	*Fundulus notatus*	3	2.8	10	13	3	2.7	14	12
Bluegill sunfish	*Lepomis macrochirus*	195	8,882	19	506	77	250	27	115
Bluntnose minnow	*Pimephales notatus*	45	77	93	102	155	200	543	294
Creek chub	*Semotilus atromaculatus*	2	1.7	16	18	307	3,356	183	1,976
Green sunfish	*Lepomis cyanellus*	40	362	9	149	189	561	179	290
Largemouth bass	*Micropterus salmoides*	36	607	17	91	-	-	1	2.9
Longear sunfish	*Lepomis megalotis*	67	1,681	37	859	19	189	32	143
Mosquitofish	*Gambusia affinis*	24	9.4	26	9	7	2.1	79	23
Quillback	*Carpiodes cyprinus*	-	-	2	14	-	-	-	-
Silverjaw minnow	*Notropis buccatus*[2]	-	-	31	102	168	292	428	372
Spotted sunfish	*Lepomis punctatus*	-	-	1	23	-	-	-	-
Steelcolor shiner	*Cyprinella whipplei*	3	7.8	-	-	-	-	-	-
Suckermouth minnow	*Phenacobius mirabilis*	62	282	71	203	-	-	-	-
White sucker	*Catostomus commersonii*	10	14	-	-	2	12	16	1,076
Yellow bullhead	*Ameiurus natalis*	27	1,137	24	445	2	19	1	101
Blackside darter	*Percina maculata*	-	-	-	-	4	11	-	-
Central stoneroller	*Campostoma anomalum*	-	-	-	-	77	376	29	135
Ribbon shiner	*Lythrurus fumerus*	-	-	-	-	1	0.6	-	-
Redfin shiner	*Lythrurus umbratilis*	-	-	-	-	14	12	20	9.7
Johnny darter	*Etheostoma nigrum*	-	-	-	-	63	59	107	93
Mississippi silvery minnow	*Hybognathus nuchalis*	-	-	-	-	-	-	3	49
Pirate perch	*Aphredoderus sayanus*	-	-	-	-	1	9.1	-	-
Creek chubsucker	*Erimyzon oblongus*	-	-	-	-	1	1.9	-	-
Total fishing distance (m)		150	-	150	-	135		135	
Total fishing time (s)		2,907	-	3,688	-	2100-2400		2100-2400	
Number of fish per site		514	-	356	-	929		1503	
Number of species per site		12	-	13	-	17	-	15	-
Total Biomass per site (g)		-	13,063	-	2,534	-	4,884	-	4,405
IBI Score			44		40		44		42

Notes:

1. Henry, D. et al. 2003. Biological Inventory and Substrate Classification in West Fork Busseron Creek, Sullian County, Indiana. Three Rivers Environmental.

2. Noted as *Ericymba buccata* in Three Rivers Environmental Report.

Table 5. West Fork Busseron Creek Fish Index of Biotic Integrity Results

Metric	ENVIRON		Three Rivers Environmental[1]	
	Site: WFBCR2 Summer 2010	Site: WFBCR3 Summer 2010	Site: "Upstream" Summer 2002	Site: "Downstream" Summer 2002
Total Number of Species	5	5	5	5
Number of Sunfish Species	3	5	3	3
Number of Sucker Species	1	1	3	1
Number of Minnow Species	3	3	5	5
Number of Sensitive Species	1	1	1	1
% Tolerant Species	5	3	3	3
% Omnivores	5	3	3	3
% Insectivores	5	5	5	5
% Pioneer Species	5	3	3	3
Catch per Unit Effort	5	5	5	5
% Simple Lithophils	1	1	3	3
% DELT Anomolies	5	5	5	5
IBI Score	**44**	**40**	**44**	**42**
Integrity Class	**Fair**	**Fair**	**Fair**	**Fair**

Notes:

1. Henry, D. et al. 2003. Biological Inventory and Substrate Classification in West Fork Busseron Creek, Sullian County, Indiana. Three Rivers Environmental.

Table 6. West Fork Busseron Creek Macroinvertebrate Survey Metric Summary for Riffle Samples Summer 2010

Benthic Macroinvertebrate Bioassesment Metric (EPA 1989)	Reference - Riffle WFBCU1		Study Sites - Riffle Samples					
			WFBCR2			WFBCR3		
	Metric Value	Score	Metric Value	% of Reference	Metric Score	Metric Value	% of Reference	Metric Score
Taxa Richness[4]	23	6	23	100	6	38	>100	6
Hilsenhoff Biotic Index (modified)[4]	5.67	6	6.65	84.8	4	7.18	79	4
Scrapers:Filter-Collectors Ratio	0.032	6	0.01	31	2	0.73	>100	6
EPT:Chironomid abundance Ratio[4]	2.22	6	0.061	2.7	0	0.79	35.4	2
Percent of Dominant Taxon[4]	47.5	2	34.1	NA[2]	2	14.5	NA[2]	6
EPT Richness[4]	2	2	3	>100	6	6	>100	6
Community Loss Index	0	6	0.69	NA[2]	4	0.37	NA[2]	6
Shredder:Total Organism Ratio	7.7	6	12.3	>100	6	20.9	>100	6
Shannon Diveristy[1]	2.91		3.11			4.04		
Total Metric Score		40			30			42
Percent of Reference Score					75.0			105.0
Biological Condition Category[3]			slight impariment from reference			no impariment from reference		

Notes:

1. Shannon Diversity is not one of the EPA 1989 scoring metrics but is included here because of the common use of this measure.
2. This metric score based on its value and not a comparison to the reference.
3. Based on USEPA 1989 Protocol III bioassessment protocols for benthic macroinvertebrate.
4. Metric also used by IDEM in mIBI determinations. Reference site metric scores adjusted by IDEM scoring criteria to reflect site-specific conditions.

Table 7. West Fork Busseron Creek Macroinvertebrate Survey Metric Summary for Multihabitat Samples Summer 2010

Benthic Macroinvertebrate Bioassesment Metric (EPA 1989)	Reference WFBCU1		Study Sites - Mulithabitat Samples					
			WFBCR2			WFBCR3		
	Metric Value	Score	Metric Value	% of Reference	Metric Score	Metric Value	% of Reference	Metric Score
Taxa Richness[4]	35	6	28	80	6	22	62.8	4
Hilsenhoff Biotic Index (modified)[4]	6.57	6	7.63	86.1	4	6.07	>100	6
Scrapers:Filter-Collectors Ratio	1.53	6	0.21	13.7	2	0.16	1.06	0
EPT:Chironomid abundance Ratio[4]	0.51	6	0.13	24.6	0	1	>100	6
Percent of Dominant Taxon[4]	19.3	6	29.3	NA[2]	2	37.5	NA[2]	2
EPT Richness[4]	3	2	4	>100	6	4	>100	6
Community Loss Index	0	6	0.89	NA[2]	4	1.22	NA[2]	4
Shredder:Total Organism Ratio	7.1	6	13.4	>100	6	16.7	>100	6
Shannon Diveristy[1]	4.13		3.64			3.26		
Total Metric Score		44			32			34
Percent of Reference Score					72.7			77.3
Biological Condition Category[3]			slight impariment from reference			slight impariment from reference		

Notes:

1. Shannon Diversity is not one of the USEPA 1989 scoring metrics but is included here because of the common use of this measure.
2. This metric score based on its value and not a comparison to the reference.
3. Based on USEPA 1989 Protocol III bioassessment protocols for benthic macroinvertebrate.
4. Metric also used by IDEM in mIBI determinations. Reference site metric scores adjusted by IDEM mIBI scoring criteria to reflect site-specific reference conditions.

Attachment 1
Habitat Metric Values

Attachment 1. Habitat Assessment Data Sheet Metric Score Summary

Metric (possible score)	Site		
	WFBCU1	WFBCR2	WFBCR3
IDEM Evaluation[1]			
Qualitative Habitat Score (total)	**53**	**52**	**50**
Substrate (20)	5	9	4
In-Stream cover (20)	6	9	8
Channel Morphology (20)	15	12	10
Riparian Zoe & Bank Erosion (10)	9	9	9
Pool/Glide Quality (12)	9	6	9
Riffle/Run Quality (8)	1	0	2
Gradient (10)	8	8	8
Percent Riffle (estimate)	5	30	20
Percent Run (estimate)	85	30	40
Percent Glide (estimate)	0	0	0
Percent Pool (estimate)	10	30	40
USEPA Evaluation[2]			
Total Score	**135**	**127**	**123**
Epifaunal Substrate/Available Cover (20)	8	3	6
Pool Substrate Characterization (20)	9	10	8
Pool Variability (20)	9	12	8
Sediment Deposition (20)	4	8	7
Channel Flow Status (20)	16	19	18
Channel Alteration (20)	19	16	16
Channel Sinuosity (20)	14	15	13
Bank Stability (LB/RB) (10/10)	9/9	7/7	8/8
Vegetative Protection (LB/RB) (10/10)	9/9	6/6	7/6
Riparian Vegetative Zone Width (LB/RB) (10/10)	10/10	9/9	9/9

Notes:

1. IDEM. 2006. Biological Studies Section, Qualitative Habitat Evaluation Index. S-001-A-BS-06-S-R1

2. USEPA. 1989. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates, and Fish, Second Ed.

Attachment 2
Fish Survey Data
Macroinvertebrate Identifications
Macroinvertebrate Metric Suite

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Blackstripe topminnow	*Fundulus notatus*	6.5	2.4		2
Blackstripe topminnow	*Fundulus notatus*	3.5	0.2		2
Blackstripe topminnow	*Fundulus notatus*	3.0	0.2		2
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.0	46.0		1
Bluegill sunfish	*Lepomis macrochirus*	15.0	66.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.3	43.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.1	52.3		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	12.7	35.5		1
Bluegill sunfish	*Lepomis macrochirus*	16.5	87.5	M	1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluegill sunfish	*Lepomis macrochirus*	15.5	75.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.8	62.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.8	56.0		1
Bluegill sunfish	*Lepomis macrochirus*	14.9	61.5		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	36.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	12.2	28.7		1
Bluegill sunfish	*Lepomis macrochirus*	15.2	64.8		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	12.5	377.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		1
Bluegill sunfish	*Lepomis macrochirus*	7.5	5.4		1
Bluegill sunfish	*Lepomis macrochirus*	7.5	5.4		1
Bluegill sunfish	*Lepomis macrochirus*	7.5	5.4		1
Bluegill sunfish	*Lepomis macrochirus*	7.5	5.4		1
Bluegill sunfish	*Lepomis macrochirus*	7.5	5.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	3.0	0.4		1
Bluegill sunfish	*Lepomis macrochirus*	16.0	79		2
Bluegill sunfish	*Lepomis macrochirus*	15.8	78		2
Bluegill sunfish	*Lepomis macrochirus*	15.5	77		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.5	47		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	38		2
Bluegill sunfish	*Lepomis macrochirus*	12.0	30		2
Bluegill sunfish	*Lepomis macrochirus*	11.5	22		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluegill sunfish	*Lepomis macrochirus*	8.0	6.4		2
Bluegill sunfish	*Lepomis macrochirus*	11.1	22		2
Bluegill sunfish	*Lepomis macrochirus*	7.5	6.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	9.0	11.0		2
Bluegill sunfish	*Lepomis macrochirus*	14.3	49		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	33		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	33		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	33		2
Bluegill sunfish	*Lepomis macrochirus*	13.0	33		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	15		2
Bluegill sunfish	*Lepomis macrochirus*	14.5	44.5		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2
Bluegill sunfish	*Lepomis macrochirus*	10.0	16.9		2
Bluegill sunfish	*Lepomis macrochirus*	8.0	7.2		2
Bluegill sunfish	*Lepomis macrochirus*	8.0	7.2		2
Bluegill sunfish	*Lepomis macrochirus*	8.0	7.2		2
Bluegill sunfish	*Lepomis macrochirus*	8.0	7.2		2
Bluegill sunfish	*Lepomis macrochirus*	7.0	6.2		2
Bluegill sunfish	*Lepomis macrochirus*	7.0	6.2		2
Bluegill sunfish	*Lepomis macrochirus*	7.0	6.2		2
Bluegill sunfish	*Lepomis macrochirus*	2.7	0.4		2
Bluegill sunfish	*Lepomis macrochirus*	2.5	0.4		2
Bluntnose minnow	*Pimephales notatus*	8.2	6.3	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	5.5	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	6.5	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	6.5	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	6.5	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	6.5	M	1
Bluntnose minnow	*Pimephales notatus*	8.2	6.5	M	1
Bluntnose minnow	*Pimephales notatus*	6.0	2.0		1
Bluntnose minnow	*Pimephales notatus*	6.0	2.0		1
Bluntnose minnow	*Pimephales notatus*	6.7	2.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluntnose minnow	*Pimephales notatus*	4.0	0.6		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.6		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.0	0.5		1
Bluntnose minnow	*Pimephales notatus*	7.7	4.3		2
Bluntnose minnow	*Pimephales notatus*	7.7	4.3		2
Bluntnose minnow	*Pimephales notatus*	7.0	1.3		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	4.4	0.6		2
Bluntnose minnow	*Pimephales notatus*	3.7	0.3		2
Bluntnose minnow	*Pimephales notatus*	4.1	0.6		2
Bluntnose minnow	*Pimephales notatus*	2.8	0.3		2
Bluntnose minnow	*Pimephales notatus*	3.0	0.2		2
Creek chub	*Semotilus atromaculatu*	4.3	0.8		1
Creek chub	*Semotilus atromaculatu*	5.5	0.9		1
Green sunfish	*Lepomis cyanellus*	12.0	27.6		1
Green sunfish	*Lepomis cyanellus*	13.0	28.2		1
Green sunfish	*Lepomis cyanellus*	12.1	27.0		1
Green sunfish	*Lepomis cyanellus*	10.5	19.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1
Green sunfish	*Lepomis cyanellus*	6.5	5.0		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Green sunfish	*Lepomis cyanellus*	17.5	7.9		1
Green sunfish	*Lepomis cyanellus*	6.0	3.5		1
Green sunfish	*Lepomis cyanellus*	7.0	4.0		1
Green sunfish	*Lepomis cyanellus*	7.0	4.0		1
Green sunfish	*Lepomis cyanellus*	7.0	4.0		1
Green sunfish	*Lepomis cyanellus*	7.0	4.0		1
Green sunfish	*Lepomis cyanellus*	7.0	4.0		1
Green sunfish	*Lepomis cyanellus*	10.0	17.0		1
Green sunfish	*Lepomis cyanellus*	4.5	1.0		1
Green sunfish	*Lepomis cyanellus*	4.5	1.0		1
Green sunfish	*Lepomis cyanellus*	4.5	1.0		1
Green sunfish	*Lepomis cyanellus*	13.5	38.8		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	8.2	11.2		2
Green sunfish	*Lepomis cyanellus*	9.5	12.4		2
Green sunfish	*Lepomis cyanellus*	7.0	5.2		2
Green sunfish	*Lepomis cyanellus*	7.0	5.2		2
Green sunfish	*Lepomis cyanellus*	7.0	5.2		2
Green sunfish	*Lepomis cyanellus*	7.0	5.2		2
Green sunfish	*Lepomis cyanellus*	8.0	9.2		2
Green sunfish	*Lepomis cyanellus*	4.3	2.0		2
Green sunfish	*Lepomis cyanellus*	4.3	2.0		2
Largemouth bass	*Micropterus salmoides*	27.0	247.0		1
Largemouth bass	*Micropterus salmoides*	16.0	45.0		1
Largemouth bass	*Micropterus salmoides*	25.0	191.0		1
Largemouth bass	*Micropterus salmoides*	17.3	59.8		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	6.3	3.0		1
Largemouth bass	*Micropterus salmoides*	6.3	3.0		1
Largemouth bass	*Micropterus salmoides*	6.3	3.0		1
Largemouth bass	*Micropterus salmoides*	6.3	3.0		1
Largemouth bass	*Micropterus salmoides*	6.0	2.4		2
Largemouth bass	*Micropterus salmoides*	6.0	2.4		2
Largemouth bass	*Micropterus salmoides*	6.0	2.4		2
Largemouth bass	*Micropterus salmoides*	6.0	2.4		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	5.5	1.8		2
Largemouth bass	*Micropterus salmoides*	7.0	4.0		2
Largemouth bass	*Micropterus salmoides*	7.0	4.0		2
Largemouth bass	*Micropterus salmoides*	7.0	4.0		2
Largemouth bass	*Micropterus salmoides*	4.2	0.8		2
Longear sunfish	*Lepomis megalotis*	13.2	41.0		1
Longear sunfish	*Lepomis megalotis*	13.6	52.0		1
Longear sunfish	*Lepomis megalotis*	13.4	48.0		1
Longear sunfish	*Lepomis megalotis*	12.7	38.0		1
Longear sunfish	*Lepomis megalotis*	12.7	36.0		1
Longear sunfish	*Lepomis megalotis*	12.0	35.0		1
Longear sunfish	*Lepomis megalotis*	12.6	47.0		1
Longear sunfish	*Lepomis megalotis*	12.6	47.0		1
Longear sunfish	*Lepomis megalotis*	12.6	47.0		1
Longear sunfish	*Lepomis megalotis*	12.6	47.0		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Longear sunfish	*Lepomis megalotis*	12.6	47.0		1
Longear sunfish	*Lepomis megalotis*	9.5	17.0		1
Longear sunfish	*Lepomis megalotis*	9.5	17.0		1
Longear sunfish	*Lepomis megalotis*	9.5	17.0		1
Longear sunfish	*Lepomis megalotis*	9.5	17.0		1
Longear sunfish	*Lepomis megalotis*	9.5	17.0		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.5	24.4		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	10.1	18.3		1
Longear sunfish	*Lepomis megalotis*	14.5	47.5		2
Longear sunfish	*Lepomis megalotis*	14.3	48.0		2
Longear sunfish	*Lepomis megalotis*	12.2	28.2		2
Longear sunfish	*Lepomis megalotis*	12.4	42.0		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Longear sunfish	*Lepomis megalotis*	12.0	35.6		2
Longear sunfish	*Lepomis megalotis*	10.5	22.4		2
Longear sunfish	*Lepomis megalotis*	10.5	22.4		2
Longear sunfish	*Lepomis megalotis*	10.5	22.4		2
Longear sunfish	*Lepomis megalotis*	10.5	22.4		2
Longear sunfish	*Lepomis megalotis*	10.5	22.4		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	10.5	22.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	8.7	12.0		2
Longear sunfish	*Lepomis megalotis*	11.0	23.5		2
Longear sunfish	*Lepomis megalotis*	8.0	9.0		2
Mosquitofish	*Gambusia affinis*	2.5	0.1	M	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6	F	1
Mosquitofish	*Gambusia affinis*	3.8	0.6		1
Mosquitofish	*Gambusia affinis*	3.8	0.6		1
Mosquitofish	*Gambusia affinis*	3.2	0.2		2
Mosquitofish	*Gambusia affinis*	4.0	0.7		2
Mosquitofish	*Gambusia affinis*	3.6	0.5		2
Mosquitofish	*Gambusia affinis*	3.8	0.5		2
Mosquitofish	*Gambusia affinis*	3.5	0.3		2
Mosquitofish	*Gambusia affinis*	2.7	0.1		2
Mosquitofish	*Gambusia affinis*	3.1	0.2		2
Mosquitofish	*Gambusia affinis*	2.1	0.1		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Mosquitofish	*Gambusia affinis*	3.3	0.3		2
Mosquitofish	*Gambusia affinis*	3.5	0.4		2
Mosquitofish	*Gambusia affinis*	3.4	0.4		2
Mosquitofish	*Gambusia affinis*	2.5	0.2		2
Mosquitofish	*Gambusia affinis*	2.5	0.2		2
Mosquitofish	*Gambusia affinis*	2.7	0.3		2
Mosquitofish	*Gambusia affinis*	2.1	0.1		2
Steelcolor shiner	*Cyprinella whipplei*	7.0	2.8		1
Steelcolor shiner	*Cyprinella whipplei*	6.5	2.2		1
Steelcolor shiner	*Cyprinella whipplei*	6.5	2.8		2
Suckermouth minnow	*Phenacobius mirabilis*	20.0	9.0		1
Suckermouth minnow	*Phenacobius mirabilis*	11.0	10.2		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	9.2	6.0		1
Suckermouth minnow	*Phenacobius mirabilis*	5.6	1.6		1
Suckermouth minnow	*Phenacobius mirabilis*	5.6	1.6		1
Suckermouth minnow	*Phenacobius mirabilis*	5.6	1.6		1
Suckermouth minnow	*Phenacobius mirabilis*	5.6	1.6		1
Suckermouth minnow	*Phenacobius mirabilis*	5.6	1.6		1
Suckermouth minnow	*Phenacobius mirabilis*	7.5	4.2		1
Suckermouth minnow	*Phenacobius mirabilis*	8.5	6.4		1
Suckermouth minnow	*Phenacobius mirabilis*	8.5	6.4		1
Suckermouth minnow	*Phenacobius mirabilis*	8.5	6.4		1
Suckermouth minnow	*Phenacobius mirabilis*	8.5	6.4		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	5.0	1.2		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	10.5	9.5		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
Suckermouth minnow	*Phenacobius mirabilis*	6.0	2.0		2
White sucker	*Catostomus commerson*	5.5	1.3		1
White sucker	*Catostomus commerson*	5.5	1.3		1
White sucker	*Catostomus commerson*	5.5	1.3		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 1 July 2010
Site: WFBCR2, 39.23606° N -87.36069°W
Reach Length: 150 meters, Shock time: 2,907 seconds

Common Name	*Genus Species*	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
White sucker	*Catostomus commerson*	5.5	1.3		1
White sucker	*Catostomus commerson*	5.5	1.3		1
White sucker	*Catostomus commerson*	5.2	1.5		2
White sucker	*Catostomus commerson*	5.2	1.5		2
White sucker	*Catostomus commerson*	5.2	1.5		2
White sucker	*Catostomus commerson*	5.2	1.5		2
White sucker	*Catostomus commerson*	5.2	1.5		2
Yellow bullhead	*Ameiurus natalis*	22.5	151.0		1
Yellow bullhead	*Ameiurus natalis*	22.0	153.0	F	1
Yellow bullhead	*Ameiurus natalis*	19.2	93.0		1
Yellow bullhead	*Ameiurus natalis*	20.0	120.0		1
Yellow bullhead	*Ameiurus natalis*	20.5	123.0		1
Yellow bullhead	*Ameiurus natalis*	17.0	61.0		1
Yellow bullhead	*Ameiurus natalis*	17.5	74.0		1
Yellow bullhead	*Ameiurus natalis*	4.8	1.0		1
Yellow bullhead	*Ameiurus natalis*	4.8	1.0		1
Yellow bullhead	*Ameiurus natalis*	4.8	1.0		1
Yellow bullhead	*Ameiurus natalis*	4.8	1.0		1
Yellow bullhead	*Ameiurus natalis*	5.0	1.5		1
Yellow bullhead	*Ameiurus natalis*	4.0	0.8		1
Yellow bullhead	*Ameiurus natalis*	24.0	188		2
Yellow bullhead	*Ameiurus natalis*	17.5	70		2
Yellow bullhead	*Ameiurus natalis*	14.5	33		2
Yellow bullhead	*Ameiurus natalis*	12.0	20		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	14.7	40		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	4.5	0.5		2
Yellow bullhead	*Ameiurus natalis*	2.5	0.5		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Blackstripe topminnow	*Fundulus notatus*	7.0	2.7		1
Blackstripe topminnow	*Fundulus notatus*	6.5	2.3		1
Blackstripe topminnow	*Fundulus notatus*	7.0	2.0		1
Blackstripe topminnow	*Fundulus notatus*	6.0	1.2		1
Blackstripe topminnow	*Fundulus notatus*	6.5	1.4		1
Blackstripe topminnow	*Fundulus notatus*	6.5	1.8		1
Blackstripe topminnow	*Fundulus notatus*	2.5	0.1		1
Blackstripe topminnow	*Fundulus notatus*	2.5	0.1		1
Blackstripe topminnow	*Fundulus notatus*	2.5	0.1		1
Blackstripe topminnow	*Fundulus notatus*	6	1.7		1
Bluegill sunfish	*Lepomis macrochirus*	16.0	68.2		1
Bluegill sunfish	*Lepomis macrochirus*	13.4	43.2		1
Bluegill sunfish	*Lepomis macrochirus*	13.9	45.0		1
Bluegill sunfish	*Lepomis macrochirus*	11.3	27.0		1
Bluegill sunfish	*Lepomis macrochirus*	15.2	62.9		1
Bluegill sunfish	*Lepomis macrochirus*	7.9	7.0		1
Bluegill sunfish	*Lepomis macrochirus*	13.2	38.2		1
Bluegill sunfish	*Lepomis macrochirus*	12.0	31.0		1
Bluegill sunfish	*Lepomis macrochirus*	10.5	19.6		1
Bluegill sunfish	*Lepomis macrochirus*	2.0	1.0		1
Bluegill sunfish	*Lepomis macrochirus*	2.4	0.1		1
Bluegill sunfish	*Lepomis macrochirus*	2.4	0.1		1
Bluegill sunfish	*Lepomis macrochirus*	2.4	0.1		1
Bluegill sunfish	*Lepomis macrochirus*	2.4	0.1		1
Bluegill sunfish	*Lepomis macrochirus*	2.4	0.1		1
Bluegill sunfish	*Lepomis macrochirus*	15.2	54.7		2
Bluegill sunfish	*Lepomis macrochirus*	12.5	33.7		2
Bluegill sunfish	*Lepomis macrochirus*	12	29.6		2
Bluegill sunfish	*Lepomis macrochirus*	13	44		2
Bluntnose minnow	*Pimephales notatus*	6.1	2.5		1
Bluntnose minnow	*Pimephales notatus*	6.5	2.6		1
bluntnose minnow	*Pimephales notatus*	7.2	3.0		1
bluntnose minnow	*Pimephales notatus*	7.8	8.0		1
bluntnose minnow	*Pimephales notatus*	6.6	2.3		1
bluntnose minnow	*Pimephales notatus*	6.4	1.2		1
bluntnose minnow	*Pimephales notatus*	6.4	1.2		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
bluntnose minnow	*Pimephales notatus*	6.4	1.2		1
bluntnose minnow	*Pimephales notatus*	6.4	1.2		1
bluntnose minnow	*Pimephales notatus*	8.3	5.0	M	1
Bluntnose minnow	*Pimephales notatus*	7.2	3.2		1
Bluntnose minnow	*Pimephales notatus*	7.2	3.2		1
Bluntnose minnow	*Pimephales notatus*	7.2	3.2		1
Bluntnose minnow	*Pimephales notatus*	7.2	3.2		1
Bluntnose minnow	*Pimephales notatus*	8	5.3		1
Bluntnose minnow	*Pimephales notatus*	6.7	3		1
Bluntnose minnow	*Pimephales notatus*	6.2	2.4		1
Bluntnose minnow	*Pimephales notatus*	6.5	2.6		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	3	0.5		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.6		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4	0.2		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4.5	0.5		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	4	0.4		1
Bluntnose minnow	*Pimephales notatus*	8.2	5.2		2
Bluntnose minnow	*Pimephales notatus*	6	*2.5*		2
Bluntnose minnow	*Pimephales notatus*	5.5	*2*		2
Bluntnose minnow	*Pimephales notatus*	6	*2.5*		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Bluntnose minnow	*Pimephales notatus*	5.5	2		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Bluntnose minnow	*Pimephales notatus*	4.2	0.63		2
Creek chub	*Semotilus atromaculatus*	5.5	1.4		1
Creek chub	*Semotilus atromaculatus*	4.5	1		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	5.1	1.3		1
Creek chub	*Semotilus atromaculatus*	4.8	0.9		1
Creek chub	*Semotilus atromaculatus*	4.8	1		1
Creek chub	*Semotilus atromaculatus*	4.8	1		1
Creek chub	*Semotilus atromaculatus*	4.8	1		1
Creek chub	*Semotilus atromaculatus*	4.8	1		1
Creek chub	*Semotilus atromaculatus*	5	1.1		2
Creek chub	*Semotilus atromaculatus*	4.5	0.95		2
Green sunfish	*Lepomis cyanellus*	13.9	49.0		1
Green sunfish	*Lepomis cyanellus*	11.8	25.5		1
Green sunfish	*Lepomis cyanellus*	10.6	19.2		1
Green sunfish	*Lepomis cyanellus*	10.1	16.5		1
Green sunfish	*Lepomis cyanellus*	8.1	8.2		1
Green sunfish	*Lepomis cyanellus*	3.0	1.0		1
Green sunfish	*Lepomis cyanellus*	3.0	1.5		1
Green sunfish	*Lepomis cyanellus*	9.8	15.5		2
Green sunfish	*Lepomis cyanellus*	9.6	12.5		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Largemouth bass	*Micropterus salmoides*	14.8	32.7		1
Largemouth bass	*Micropterus salmoides*	7.9	5.2		1
Largemouth bass	*Micropterus salmoides*	8.5	7.0		1
Largemouth bass	*Micropterus salmoides*	5.8	2.7		1
Largemouth bass	*Micropterus salmoides*	7.5	5.6		1
Largemouth bass	*Micropterus salmoides*	6.5	3.8		1
Largemouth bass	*Micropterus salmoides*	6.0	2.4		1
Largemouth bass	*Micropterus salmoides*	7.0	4.8		1
Largemouth bass	*Micropterus salmoides*	5.8	3.0		1
Largemouth bass	*Micropterus salmoides*	6.2	4.2		1
Largemouth bass	*Micropterus salmoides*	6.0	3.6		1
Largemouth bass	*Micropterus salmoides*	7.1	3.9		1
Largemouth bass	*Micropterus salmoides*	6.0	1.2		1
Largemouth bass	*Micropterus salmoides*	8.0	5.0		1
Largemouth bass	*Micropterus salmoides*	7.0	3.3		1
Largemouth bass	*Micropterus salmoides*	5.2	1.6		1
Largemouth bass	*Micropterus salmoides*	4.7	0.6		1
Longear sunfish	*Lepomis megalotis*	10.0	22.0		1
Longear sunfish	*Lepomis megalotis*	11.0	28.7		1
Longear sunfish	*Lepomis megalotis*	12.2	39.4		1
Longear sunfish	*Lepomis megalotis*	9.7	1.7		1
Longear sunfish	*Lepomis megalotis*	14.1	58.6		1
Longear sunfish	*Lepomis megalotis*	12.9	46.0		1
Longear sunfish	*Lepomis megalotis*	10.0	21.0		1
Longear sunfish	*Lepomis megalotis*	10.9	24.5		1
Longear sunfish	*Lepomis megalotis*	10.0	21.4		1
Longear sunfish	*Lepomis megalotis*	9.5	18.0		1
Longear sunfish	*Lepomis megalotis*	11.2	28.2		1
Longear sunfish	*Lepomis megalotis*	13.2	40.0		1
Longear sunfish	*Lepomis megalotis*	9.6	17.3		1
Longear sunfish	*Lepomis megalotis*	9.6	17.0		1
Longear sunfish	*Lepomis megalotis*	11.0	23.0		1
Longear sunfish	*Lepomis megalotis*	12.0	34.5		1
Longear sunfish	*Lepomis megalotis*	10.4	22.3		1
Longear sunfish	*Lepomis megalotis*	10.0	18.5		1
Longear sunfish	*Lepomis megalotis*	9.6	16.0		1
Longear sunfish	*Lepomis megalotis*	10.7	19.9		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Longear sunfish	*Lepomis megalotis*	7.5	6.7		1
Longear sunfish	*Lepomis megalotis*	11.2	23.5		1
Longear sunfish	*Lepomis megalotis*	10.0	17.9		1
Longear sunfish	*Lepomis megalotis*	11.3	29.0		1
Longear sunfish	*Lepomis megalotis*	10.3	22.7		1
Longear sunfish	*Lepomis megalotis*	9.5	16.2		1
Longear sunfish	*Lepomis megalotis*	15.2	58		2
Longear sunfish	*Lepomis megalotis*	12	33.4		2
Longear sunfish	*Lepomis megalotis*	9.8	19.1		2
Longear sunfish	*Lepomis megalotis*	11	23.9		2
Longear sunfish	*Lepomis megalotis*	10	22.9		2
Longear sunfish	*Lepomis megalotis*	12.8	40.5		2
Longear sunfish	*Lepomis megalotis*	9.3	15.4		2
Longear sunfish	*Lepomis megalotis*	7.5	4.3		2
Longear sunfish	*Lepomis megalotis*	7.5	4.3		2
Longear sunfish	*Lepomis megalotis*	5.5	1.7		2
Longear sunfish	*Lepomis megalotis*	5.5	1.7		2
Mosquitofish	*Gambusia affinis*	4.0	1.0		1
Mosquitofish	*Gambusia affinis*	4.7	1.5		1
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	2.5	0.1		1
Mosquitofish	*Gambusia affinis*	3	0.5		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	3	0.5		1
Mosquitofish	*Gambusia affinis*	5.2	*1.2*		2
Mosquitofish	*Gambusia affinis*	3.5	*0.54*		2
Quillback	*Carpiodes cyprinus*	7.9	6.2		1
Quillback	*Carpiodes cyprinus*	8.5	7.8		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	8.1	5.2		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	7.4	3.0		1
Silverjaw minnow	*Notropis buccatus*	8.0	3.5		1
Silverjaw minnow	*Notropis buccatus*	8.0	3.5		1
Silverjaw minnow	*Notropis buccatus*	8.0	3.5		1
Silverjaw minnow	*Notropis buccatus*	4.8	1		1
Silverjaw minnow	*Notropis buccatus*	4.8	1		1
Silverjaw minnow	*Notropis buccatus*	4.8	1		1
Silverjaw minnow	*Notropis buccatus*	4.8	1		1
Silverjaw minnow	*Notropis buccatus*	4.8	1		1
Silverjaw minnow	*Notropis buccatus*	8	3.8		2
Silverjaw minnow	*Notropis buccatus*	4.8	0.8		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Silverjaw minnow	*Notropis buccatus*	4.5	*0.41*		2
Silverjaw minnow	*Notropis buccatus*	4.5	*0.41*		2
Spotted sunfish	*Lepomis punctatus*	10.9	22.7		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	9.0	6.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	1		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	5	0.8		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	4.8	0.9		1
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	5.2	1.8		2
Suckermouth minnow	*Phenacobius mirabilis*	3.2	*0.3*		2
Suckermouth minnow	*Phenacobius mirabilis*	6	*2.5*		2
Suckermouth minnow	*Phenacobius mirabilis*	9.5	7.8		2
Suckermouth minnow	*Phenacobius mirabilis*	9.5	7.8		2
Suckermouth minnow	*Phenacobius mirabilis*	9.5	7.8		2
Suckermouth minnow	*Phenacobius mirabilis*	9.5	7.8		2

West Fork Busseron Creek Fish Survey Results

Sample Date: 30 June 2010
Site: WFBCR3, 39.23132° N -87.35915°W
Reach Length: 150 meters, Shock time: 3,688 seconds

Common Name	Genus Species	Total Length (cm)	Weight (g)	Sex (M/F)	Pass No.
Yellow bullhead	*Ameiurus natalis*	18.5	89		1
Yellow bullhead	*Ameiurus natalis*	17	66		1
Yellow bullhead	*Ameiurus natalis*	14	33.5		1
Yellow bullhead	*Ameiurus natalis*	15	40		1
Yellow bullhead	*Ameiurus natalis*	12.5	25.2		1
Yellow bullhead	*Ameiurus natalis*	5.3	1.0		1
Yellow bullhead	*Ameiurus natalis*	11.0	18.8		1
Yellow bullhead	*Ameiurus natalis*	5.5	3.0		1
Yellow bullhead	*Ameiurus natalis*	4.5	1.7		1
Yellow bullhead	*Ameiurus natalis*	3.2	1.2		1
Yellow bullhead	*Ameiurus natalis*	4.2	1.6		1
Yellow bullhead	*Ameiurus natalis*	3.0	0.6		1
Yellow bullhead	*Ameiurus natalis*	4.3	1.0		1
Yellow bullhead	*Ameiurus natalis*	4.0	1.4		1
Yellow bullhead	*Ameiurus natalis*	2.5	0.1		1
Yellow bullhead	*Ameiurus natalis*	4.6	0.5		1
Yellow bullhead	*Ameiurus natalis*	4.8	2.0		1
Yellow bullhead	*Ameiurus natalis*	11	18.2		2
Yellow bullhead	*Ameiurus natalis*	12	20.5		2
Yellow bullhead	*Ameiurus natalis*	12	24.4		2
Yellow bullhead	*Ameiurus natalis*	18	19		2
Yellow bullhead	*Ameiurus natalis*	18	74.6		2
Yellow bullhead	*Ameiurus natalis*	4.5	1		2
Yellow bullhead	*Ameiurus natalis*	3.5	0.7		1

1. Values in italics were estimated based on weight/length relationship.

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are NOT adjusted for subsampling

	Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
	Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
	Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
	Device	**3m2**	**3m2**	**3m2**
	Habitat	**Riffle**	**Riffle**	**Riffle**
	Percent Subsampled	**100.00**	**6.25**	**22.94**
	EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
Ephemeroptera	Baetidae	0	0	1
	Caenis sp.	1	1	8
	Tricorythodes sp.	0	0	1
Odonata	Argia sp.	0	0	1
	Coenagrionidae	1	0	40
Hemiptera	Corixidae	0	0	1
Coleoptera	Berosus sp.	0	2	43
	Coptotomus sp.	0	0	1
	Dubiraphia sp.	1	0	0
	Peltodytes sp.	0	0	3
	Tropisternus sp.	0	0	1
Megaloptera	Sialis sp.	2	0	0
Diptera-Chironomidae	Ablabesmyia mallochi	0	0	3
	Cladotanytarsus sp.	1	0	0
	Cricotopus bicinctus gr.	0	25	3
	Cricotopus sp.	0	0	2
	Cryptochironomus sp.	9	8	0
	Dicrotendipes neomodestus	4	36	21
	Dicrotendipes simpsoni	0	1	2
	Endochironomus sp.	0	1	2
	Glyptotendipes sp.	0	45	17
	Harnischia sp.	0	0	1
	Labrundinia sp.	0	0	1
	Parachironomus sp.	0	1	3
	Polypedilum flavum	10	6	8
	Polypedilum halterale gr.	0	1	0

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are NOT adjusted for subsampling

EcoAnalysts, Inc.

		W.F. Busseron Crk	W.F. Busseron Crk	W.F. Busseron Crk
	Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
	Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
	Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
	Device	**3m2**	**3m2**	**3m2**
	Habitat	**Riffle**	**Riffle**	**Riffle**
	Percent Subsampled	**100.00**	**6.25**	**22.94**
	EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
	Polypedilum scalaenum gr.	1	1	0
	Pseudochironomus sp.	0	97	6
	Rheotanytarsus exiguus gr.	0	21	7
	Saetheria tylus	1	0	0
	Stictochironomus sp.	1	0	0
	Thienemannimyia gr. sp.	4	1	4
Diptera	Bezzia/Palpomyia sp.	8	0	0
	Dolichopodidae	0	0	1
	Erioptera sp.	0	1	0
	Hedriodiscus/Odontomyia sp.	1	0	1
	Sciomyzidae	0	0	1
	Simulium sp.	0	0	2
	Tabanidae	1	0	0
	Tipulidae	0	0	1
Trichoptera	Cheumatopsyche sp.	68	10	12
	Hydroptila sp.	0	4	40
	Oxyethira sp.	0	0	1
Gastropoda	Fossaria sp.	2	0	10
	Helisoma anceps	1	0	0
	Physa sp.	0	0	40
	Planorbidae	0	2	0
Bivalvia	Sphaeriidae	19	0	0
	Utterbackia sp.	1	0	0
Annelida	Enchytraeidae	0	2	0
	Helobdella sp.	1	0	0
	Limnodrilus hoffmeisteri	0	16	0

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are NOT adjusted for subsampling

EcoAnalysts, Inc.

	Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
	Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
	Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
	Device	**3m2**	**3m2**	**3m2**
	Habitat	**Riffle**	**Riffle**	**Riffle**
	Percent Subsampled	**100.00**	**6.25**	**22.94**
	EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
	Pristina jenkinae	0	1	0
	Tubificidae w/o cap setae	0	0	2
Acari	Koenikea sp.	0	0	1
Crustacea	Cambaridae	2	0	0
	Hyalella sp.	0	0	3
	Orconectes sp.	0	0	1
Other Organisms	Nematoda	3	0	0
	Prostoma sp.	0	1	0
	TOTAL	**143**	**284**	**296**

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	W.F. Busseron Crk	W.F. Busseron Crk	W.F. Busseron Crk
Site	WFBCU1	WFBCR2	WFBCR3
Date	06-29-2010	06-29-2010	06-29-2010
Device	3m2	3m2	3m2
Habitat	Riffle	Riffle	Riffle
Percent Subsampled	100.00	6.25	22.94
EcoAnalysts Sample ID	5508.1-1	5508.1-2	5508.1-3
Abundance Measures			
Corrected Abundance	143.00	4544.00	1290.56
EPT Abundance	69.00	240.00	274.68
Dominance Measures			
Dominant Taxon	Cheumatopsyche sp.	Pseudochironomus sp.	Berosus sp.
Dominant Abundance	68.00	1552.00	187.48
2nd Dominant Taxon	Sphaeriidae	Glyptotendipes sp.	Physa sp.
2nd Dominant Abundance	19.00	720.00	174.40
3rd Dominant Taxon	Polypedilum flavum	Dicrotendipes neomodestus	Coenagrionidae
3rd Dominant Abundance	10.00	576.00	174.40
% Dominant Taxon	47.55	34.15	14.53
% 2 Dominant Taxa	60.84	50.00	28.04
% 3 Dominant Taxa	67.83	62.68	41.55
Richness Measures			
Species Richness	23.00	23.00	38.00
EPT Richness	2.00	3.00	6.00
Ephemeroptera Richness	1.00	1.00	3.00
Plecoptera Richness	0.00	0.00	0.00
Trichoptera Richness	1.00	2.00	3.00
Chironomidae Richness	8.00	13.00	14.00
Oligochaeta Richness	0.00	3.00	1.00
Non-Chiro. Non-Olig. Richness	15.00	7.00	23.00

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Riffle**	**Riffle**	**Riffle**
Percent Subsampled	**100.00**	**6.25**	**22.94**
EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
Rhyacophila Richness	0.00	0.00	0.00
Community Composition			
% Ephemeroptera	0.70	0.35	3.38
% Plecoptera	0.00	0.00	0.00
% Trichoptera	47.55	4.93	17.91
% EPT	48.25	5.28	21.28
% Coleoptera	0.70	0.70	16.22
% Diptera	28.67	86.27	29.05
% Oligochaeta	0.00	6.69	0.68
% Baetidae	0.00	0.00	0.34
% Brachycentridae	0.00	0.00	0.00
% Chironomidae	21.68	85.92	27.03
% Ephemerellidae	0.00	0.00	0.00
% Hydropsychidae	47.55	3.52	4.05
% Odonata	0.70	0.00	13.85
% Perlidae	0.00	0.00	0.00
% Pteronarcyidae	0.00	0.00	0.00
% Simuliidae	0.00	0.00	0.68
Functional Group Composition			
% Filterers	60.84	10.92	7.09
% Gatherers	4.90	58.10	15.54
% Predators	19.58	3.52	17.23
% Scrapers	2.10	0.70	16.89

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are adjusted for subsampling

ECOANALYSTS, INC.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Riffle**	**Riffle**	**Riffle**
Percent Subsampled	**100.00**	**6.25**	**22.94**
EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
% Shredders	7.69	12.32	20.95
% Piercer-Herbivores	0.00	1.41	14.19
% Unclassified	4.90	13.03	8.11
Filterer Richness	2.00	2.00	3.00
Gatherer Richness	6.00	9.00	12.00
Predator Richness	7.00	3.00	7.00
Scraper Richness	2.00	1.00	2.00
Shredder Richness	2.00	5.00	7.00
Piercer-Herbivore Richness	0.00	1.00	3.00
Unclassified	4.00	2.00	4.00
Diversity/Evenness Measures			
Shannon-Weaver H' (log 10)	0.88	0.93	1.22
Shannon-Weaver H' (log 2)	2.91	3.11	4.04
Shannon-Weaver H' (log e)	2.02	2.15	2.80
Margalef's Richness	4.43	2.61	5.17
Pielou's J'	0.64	0.69	0.77
Simpson's Heterogeneity	0.75	0.82	0.91
Biotic Indices			
% Indiv. w/ HBI Value	90.91	99.65	98.65
Hilsenhoff Biotic Index	5.67	6.65	7.18
% Indiv. w/ MTI Value	78.32	44.72	44.93
Metals Tolerance Index	4.38	4.02	3.74
% Indiv. w/ FSBI Value	48.95	4.93	18.58

ENVIRON IN Stream Restoration Benthos 2010 (Riffles)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Riffle**	**Riffle**	**Riffle**
Percent Subsampled	**100.00**	**6.25**	**22.94**
EcoAnalysts Sample ID	**5508.1-1**	**5508.1-2**	**5508.1-3**
Fine Sediment Biotic Index	3.00	7.00	14.00
FSBI - average	0.13	0.30	0.37
FSBI - weighted average	1.97	2.86	4.25
% Indiv. w/ TPM Value	60.14	23.94	28.38
Temp. Pref. Metric - average	0.39	0.57	0.74
TPM - weighted average	1.17	1.79	2.04
Other Metrics			
Long-Lived Taxa Richness	2.00	0.00	1.00
Clinger Richness	6.00	7.00	16.00
% Clingers	52.45	38.03	48.31
Intolerant Taxa Richness	0.00	0.00	0.00
% Tolerant Individuals	1.54	0.44	6.76
% Tolerant Taxa	21.74	43.48	31.58
Coleoptera Richness	1.00	1.00	4.00

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are NOT adjusted for subsampling

EcoAnalysts, Inc.

	Water Body	W.F. Busseron Crk	W.F. Busseron Crk	W.F. Busseron Crk
	Site	WFBCU1	WFBCR2	WFBCR3
	Date	06-29-2010	06-29-2010	06-29-2010
	Device	3m2	3m2	3m2
	Habitat	Debris	Debris	Debris
	Percent Subsampled	100.00	12.50	7.29
	EcoAnalysts Sample ID	5508.2-1	5508.2-2	5508.2-3
Ephemeroptera	Baetis intercalaris	0	0	8
	Caenis sp.	4	1	1
Odonata	Argia sp.	1	0	0
	Boyeria vinosa	1	0	0
	Coenagrionidae	6	26	0
	Corduliidae	2	0	0
Coleoptera	Berosus sp.	0	16	11
	Dubiraphia sp.	3	0	0
	Enochrus sp.	0	1	0
	Helichus sp.	1	0	0
	Macronychus glabratus	1	0	0
	Neoporus sp.	1	0	0
	Peltodytes sp.	4	0	0
Diptera-Chironomidae	Ablabesmyia mallochi	4	1	0
	Chironomus sp.	1	0	0
	Clinotanypus sp.	1	0	0
	Cricotopus bicinctus gr.	0	1	3
	Cricotopus sp.	0	2	2
	Cryptochironomus sp.	1	0	10
	Cryptotendipes sp.	1	0	0
	Dicrotendipes neomodestus	1	39	21
	Dicrotendipes simpsoni	0	2	0
	Endochironomus sp.	0	6	0
	Glyptotendipes sp.	0	81	12
	Parachironomus sp.	0	11	0
	Paratendipes sp.	1	0	0

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are NOT adjusted for subsampling

ECOANALYSTS, INC.

	Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
	Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
	Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
	Device	**3m2**	**3m2**	**3m2**
	Habitat	**Debris**	**Debris**	**Debris**
	Percent Subsampled	**100.00**	**12.50**	**7.29**
	EcoAnalysts Sample ID	**5508.2-1**	**5508.2-2**	**5508.2-3**
	Phaenopsectra sp.	1	0	0
	Polypedilum flavum	0	10	31
	Polypedilum halterale gr.	0	0	2
	Polypedilum illinoense gr.	3	2	0
	Procladius sp.	2	0	0
	Pseudochironomus sp.	0	7	28
	Rheotanytarsus exiguus gr.	0	13	11
	Thienemannimyia gr. sp.	2	0	0
	Xenochironomus xenolabis	3	0	0
Diptera	Bezzia/Palpomyia sp.	5	1	1
	Ceratopogoninae	0	1	0
	Erioptera sp.	0	0	2
	Stratiomyidae	1	0	0
Trichoptera	Cheumatopsyche sp.	6	13	105
	Hydroptila sp.	1	7	7
	Oecetis sp.	0	1	0
Lepidoptera	Lepidoptera	1	0	0
Gastropoda	Fossaria sp.	11	5	2
	Gyraulus sp.	0	7	0
	Helisoma anceps	27	0	0
	Physa sp.	19	16	1
Bivalvia	Musculium sp.	0	0	1
	Pisidium sp.	20	0	0
Annelida	Erpobdella sp.	0	1	6
	Glossiphoniidae	1	0	0
	Lumbricina	1	0	0

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are NOT adjusted for subsampling

EcoAnalysts, Inc.

		Water Body	W.F. Busseron Crk	W.F. Busseron Crk	W.F. Busseron Crk
		Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
		Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
		Device	**3m2**	**3m2**	**3m2**
		Habitat	**Debris**	**Debris**	**Debris**
		Percent Subsampled	**100.00**	**12.50**	**7.29**
		EcoAnalysts Sample ID	**5508.2-1**	**5508.2-2**	**5508.2-3**
	Tubificidae w/o cap setae		0	0	14
Acari	Mideopsis sp.		1	0	0
	Neumania sp.		1	0	0
Crustacea	Cambaridae		0	2	1
	Hyalella sp.		0	1	0
	Ostracoda		0	2	0
		TOTAL	**140**	**276**	**280**

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Debris**	**Debris**	**Debris**
Percent Subsampled	**100.00**	**12.50**	**7.29**
EcoAnalysts Sample ID	**5508.2-1**	**5508.2-2**	**5508.2-3**
Abundance Measures			
Corrected Abundance	140.00	2208.00	3838.80
EPT Abundance	11.00	176.00	1658.91
Dominance Measures			
Dominant Taxon	Helisoma anceps	Glyptotendipes sp.	Cheumatopsyche sp.
Dominant Abundance	27.00	648.00	1439.55
2nd Dominant Taxon	Pisidium sp.	Dicrotendipes neomodestus	Polypedilum flavum
2nd Dominant Abundance	20.00	312.00	425.01
3rd Dominant Taxon	Physa sp.	Coenagrionidae	Pseudochironomus sp.
3rd Dominant Abundance	19.00	208.00	383.88
% Dominant Taxon	19.29	29.35	37.50
% 2 Dominant Taxa	33.57	43.48	48.57
% 3 Dominant Taxa	47.14	52.90	58.57
Richness Measures			
Species Richness	35.00	28.00	22.00
EPT Richness	3.00	4.00	4.00
Ephemeroptera Richness	1.00	1.00	2.00
Plecoptera Richness	0.00	0.00	0.00
Trichoptera Richness	2.00	3.00	2.00
Chironomidae Richness	12.00	12.00	9.00
Oligochaeta Richness	1.00	0.00	1.00
Non-Chiro. Non-Olig. Richness	22.00	16.00	12.00

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are adjusted for subsampling

ECOANALYSTS, INC.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Debris**	**Debris**	**Debris**
Percent Subsampled	**100.00**	**12.50**	**7.29**
EcoAnalysts Sample ID	**5508.2-1**	**5508.2-2**	**5508.2-3**
Rhyacophila Richness	0.00	0.00	0.00
Community Composition			
% Ephemeroptera	2.86	0.36	3.21
% Plecoptera	0.00	0.00	0.00
% Trichoptera	5.00	7.61	40.00
% EPT	7.86	7.97	43.21
% Coleoptera	7.14	6.16	3.93
% Diptera	19.29	64.13	43.93
% Oligochaeta	0.71	0.00	5.00
% Baetidae	0.00	0.00	2.86
% Brachycentridae	0.00	0.00	0.00
% Chironomidae	15.00	63.41	42.86
% Ephemerellidae	0.00	0.00	0.00
% Hydropsychidae	4.29	4.71	37.50
% Odonata	7.14	9.42	0.00
% Perlidae	0.00	0.00	0.00
% Pteronarcyidae	0.00	0.00	0.00
% Simuliidae	0.00	0.00	0.00
Functional Group Composition			
% Filterers	18.57	9.42	41.79
% Gatherers	8.57	38.77	23.57
% Predators	21.43	10.87	3.93
% Scrapers	41.43	10.14	1.07

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	W.F. Busseron Crk	W.F. Busseron Crk	W.F. Busseron Crk
Site	WFBCU1	WFBCR2	WFBCR3
Date	06-29-2010	06-29-2010	06-29-2010
Device	3m2	3m2	3m2
Habitat	Debris	Debris	Debris
Percent Subsampled	100.00	12.50	7.29
EcoAnalysts Sample ID	5508.2-1	5508.2-2	5508.2-3
% Shredders	7.14	13.41	16.79
% Piercer-Herbivores	0.71	2.54	2.50
% Unclassified	2.14	14.86	10.36
Filterer Richness	2.00	2.00	3.00
Gatherer Richness	7.00	8.00	7.00
Predator Richness	14.00	5.00	2.00
Scraper Richness	4.00	3.00	2.00
Shredder Richness	5.00	6.00	4.00
Piercer-Herbivore Richness	1.00	1.00	1.00
Unclassified	2.00	3.00	3.00
Diversity/Evenness Measures			
Shannon-Weaver H' (log 10)	1.24	1.10	0.98
Shannon-Weaver H' (log 2)	4.13	3.64	3.26
Shannon-Weaver H' (log e)	2.86	2.52	2.26
Margalef's Richness	6.88	3.51	2.54
Pielou's J'	0.80	0.76	0.73
Simpson's Heterogeneity	0.91	0.87	0.82
Biotic Indices			
% Indiv. w/ HBI Value	72.86	98.91	96.79
Hilsenhoff Biotic Index	6.57	7.63	6.07
% Indiv. w/ MTI Value	29.29	31.52	55.36
Metals Tolerance Index	3.51	3.94	4.72
% Indiv. w/ FSBI Value	5.00	7.25	40.00

ENVIRON IN Stream Restoration Benthos 2010 (Multi Habitats)
Data are adjusted for subsampling

EcoAnalysts, Inc.

Water Body	**W.F. Busseron Crk**	**W.F. Busseron Crk**	**W.F. Busseron Crk**
Site	**WFBCU1**	**WFBCR2**	**WFBCR3**
Date	**06-29-2010**	**06-29-2010**	**06-29-2010**
Device	**3m2**	**3m2**	**3m2**
Habitat	**Debris**	**Debris**	**Debris**
Percent Subsampled	**100.00**	**12.50**	**7.29**
EcoAnalysts Sample ID	**5508.2-1**	**5508.2-2**	**5508.2-3**
Fine Sediment Biotic Index	7.00	7.00	7.00
FSBI - average	0.20	0.25	0.32
FSBI - weighted average	2.43	3.05	2.19
% Indiv. w/ TPM Value	11.43	17.75	56.79
Temp. Pref. Metric - average	0.29	0.64	0.64
TPM - weighted average	1.44	2.10	1.43
Other Metrics			
Long-Lived Taxa Richness	3.00	0.00	1.00
Clinger Richness	9.00	11.00	9.00
% Clingers	52.14	55.07	53.93
Intolerant Taxa Richness	1.00	0.00	0.00
% Tolerant Individuals	27.45	2.43	0.40
% Tolerant Taxa	34.29	42.86	22.73
Coleoptera Richness	5.00	2.00	1.00

Attachment 3
West Fork Busseron Creek
Study Site Photographs

Attachment 3. West Fork Busseron Creek Fish and Macroinvertebrate Survey June 29-July 1, 2010

Figure 1. Site WFBCU1 riffle area looking upstream. Macroinvertebrate sampling area.
West Fork Busseron Creek near County Road 2125N. June 29, 2010.

Figure 2. Site WFBCU1 riffle macroinvertebrate sampling. West Fork Busseron Creek near County Road 2124N. June 29, 2010.

Figure 3. Site WFBCR2 looking downstream. West Fork Busseron Creek within upper portion of mitigation area WFBCM. June 29, 2010.

Figure 4. Site WFBCR2 example riffle area. Macroinvertebrate dip net sampling. West Fork Busseron Creek mitigation area WFBCM. June 29, 2010.

Figure 5. Site WFBCR2. Near downstream end of study reach looking upstream. West Fork Busseron Creek mitigation area WFBCM. June 29, 2010.

Figure 6. Site WFBCR3. Middle of study reach looking upstream. West Fork Busseron Creek mitigation area WFBCM. June 29, 2010.

Attachment 3. West Fork Busseron Creek Fish and Macroinvertebrate Survey June 29-July 1, 2010

Figure 7. Site WFBCR3. Downstream end of study reach looking upstream. West Fork Busseron Creek mitigation area WFBCM. June 29, 2010.

Figure 8. Site WFBCR3. Macroinvertebrate vegetation/debris dam habitat sampling. West Fork Busseron Creek mitigation area WFBCM. June 29, 2010.

Attachment 3. West Fork Busseron Creek Fish and Macroinvertebrate Survey June 29-July 1, 2010

Figure 9. Site WFBCR2. Fish survey sunfish example. West Fork Busseron Creek mitigation area WFBCM. July 1, 2010.

Figure 10. Site WFBCR2. Fish survey largemouth bass example. West Fork Busseron Creek mitigation area WFBCM. July 1, 2010.

Figure 11. Site WFBCR2. Electrofishing West Fork Busseron Creek. Undercut bank habitat with many sunfish. July 1, 2010.

Figure 12. Site WFBCR2. Cattail bed habitat. Site for many sunfish, bass, and catfish specimens captured during electrofishing. West Fork Busseron Creek within mitigation area WFBCM. July 1, 2010.

APPENDIX B – EXHIBIT 5

Freshwater Mussel Survey Results
West Fork Busseron Creek Mitigation Area
Farmersburg, Indiana

Prepared for:

Peabody Energy
Evansville, Indiana

Prepared by:

ENVIRON International Corporation
Denver, Colorado

Date:
August 30, 2011

ENVIRON

Contents

Executive Summary

A freshwater mussel survey was conducted by ENVIRON International Corporation (ENVIRON) within the West Fork Busseron Creek Mitigation Area (WFBCMA) near Farmersburg, Sullivan County, Indiana. The WFBCMA is a post-mining reclamation action completed in 2006 by Peabody Midwest Mining, LLC, in the West Fork Busseron Creek. Pre-mining bioassessment results indicated no mussels to be present in this portion of West Fork Busseron Creek. However, during post-reclamation bioassessment monitoring of fish and benthic macroinvertebrate, specimens of freshwater mussels were observed in the WFBCMA. This report presents the results of a mussel survey conducted on August 5, 2011 to characterize and evaluate freshwater mussels in the WFBCMA.

The mussel survey in the WFBCMA used standard mussel survey methods based on systematic sampling from randomly selected survey plots associated with a stratified distribution of cross-stream transects in a stream study reach. The WFBCMA survey incorporated two study stream reaches a minimum of 150 feet in length located near the upstream and downstream portions of the mitigation area. Stream sediment from within a 0.25 M^2 survey plot was removed to a depth of approximately 4-5 inches and sieved through a ¼ inch screen. All mussels encountered were identified to species, measured (total length), and counted prior to replacement in the stream. Key findings of the mussel survey in the WFBCMA include the following:

1. *Ligumia subrostrata* (Pondmussel) and *Utterbackia imbecillis* (Paper Pondmussel) are present in the upper portion of the WFBCMA.
2. A total of 39 stream survey plots were evaluated for mussels. Extrapolated total mussel density was 3.4 mussels/M^2 with *Ligumia subrostrata* density being double that of *Utterbackia imbecillis*.
3. Analysis of annular shell growth rings and comparison to length measurements suggest that mussels have been colonizing the upper portion of the WFBCMA for at least two to three years. Maximum valve length was 85.3 mm for *U. imbecillis* and 33.7 mm for *L. subrostrata*.
4. The downstream study reach was positioned below a recently constructed and active beaver dam. No mussels were encountered in any of 10 stream survey plots from below the beaver dam. Construction of the beaver dam and recent deposition in the streambed from an episodic event, each occurring after the 2010 bioassessment, may have contributed to the lack of mussels in this portion of the WFBCMA.
5. Mean field measured water quality parameters or pH (7.5 su), dissolved oxygen (8.5 mg/L), temperature (32 °C), and specific conductivity (275 mS/cm) indicated warm water conditions consistent with the geographical area. However, the lowest dissolved oxygen concentration and a lowering of water temperature appeared to a consequence of the beaver dam.

Given the complex life cycle and requirements for freshwater mussels to occur in streams, the presence of multiple Unionid mussel species in the WFBCMA is further evidence that biological function of the stream has been attained.

Introduction

ENVIRON International Corporation (ENVIRON) conducted a freshwater mussel survey within the West Fork Busseron Creek Mitigation Area (WFBMA), Farmersburg, Sullivan County, Indiana. Peabody Midwest Mining, LLC (Peabody) established the WFBMA as reclamation following of mining activities at the Farmersburg Mine. Bioassessment of the West Fork Busseron Creek prior to mining disturbance included a formal mussel survey, from which no mussel specimens were reported (Three Rivers 2003 *Biological Inventory and Substrate Classifications in West Fork Busseron Creek, Sullivan County Indiana*). The WFBCM area is approximately 7,825 feet in length and stream reconstruction plans incorporated current aspects and understanding of hydrology and stream morphology to enhance the ecological benefits of the stream specific to the gradient and geographical area.

Four years following reclamation, ENVIRON conducted a benthic macroinvertebrate and fish bioassessment in the WFBCMA (June 29-July 1, 2010). The bioassessment provided biological information as a temporal benchmark for demonstrating community composition and functional aspects of the stream which were equivalent with pre-mining conditions. During this bioassessment, ENVIRON incidentally observed the presence of Unionid freshwater mussels within in the study area.

This 2011 mussel survey was conducted to verify and quantify the presence of freshwater mussels in the WFBMA following Peabody Energy's reclamation actions. The distribution and abundance of freshwater mussels has been a recent focus of environmental and ecological concern of state, federal, and special interest groups. Documentation of a freshwater mussel population in the WFBCMA is a strong ecological statement that further demonstrates successful mitigation resulting in good water quality, and restoration of stream function.

Methods

Water Quality

Water quality measurements were recorded in situ using a portable Horiba water quality multi-probe meter. Parameters, including pH (s.u), dissolved oxygen (mg/L), conductivity (µmhos/cm), and temperature (°C), were taken intermittently during the day to account for any diurnal fluctuation. No water samples were collected for other chemical parameters.

Instantaneous discharge was determined at the survey reference locations. Total discharge was calculated by the incremental flow method using a standard top-setting rod and Marsh-McBirney Model 2000 velocity meter to obtain depth and water velocity data at numerous intervals across the stream.

Mussel Survey

ENVIRON staff conducted a mussel survey with assistance from Peabody Energy personnel in the WFBCMA on August 5, 2011. The survey incorporated two stream reaches, one positioned in the upper portion of the mitigation area and the other near the downstream portion of WFBC mitigation area (Figure 1). The survey followed standard techniques of Strayer and Smith (2003) and used systematic sampling from random survey plots selected within a stratified distribution of transects. At each study reach, a reference location was selected, from which numerous cross-stream transects were established at uniform intervals in an upstream or downstream direction. At each cross-stream transect, a 0.25 M^2 sampling frame was positioned in the deepest portion of the stream flow (typically the center) to identify the sampling area (see Photo 5).

Mussel Survey Study Reaches

Figure 1 shows the location of the mussel survey study reaches in the WFBCMA. The reference location for the upper WFBCMA survey reach was latitude 39° 14.124' N and longitude -87° 21.616' W. Mussel sampling transects were positioned every 15 feet (ft) upstream of this location for a distance of 300 ft (see Photos 10 and 11). Due to mussel survey sampling results in the lower study area that was limited by the presence of a beaver dam, the upper mussel survey area was extended, and sampling transects were also positioned every 20 ft downstream of the reference location for a distance of 150 ft (see Photo 12). Thus, 29 sample plots for a total area of 7.25 M^2 were surveyed for mussels in the upper survey reach of the WFBCMA.

The reference location for the lower WFBCMA survey reach was latitude 39° 13.650' N and longitude -87° 21.434' W. This location is downstream of a beaver dam established since the 2010 bioassessment study and the location selected to best match flow conditions, stream width, and general gradient of the upper mussel survey area. Mussel sampling transects were positioned every 15 ft upstream of the reference location for a distance of 150 ft towards the beaver dam (Photo 16).

Mussel Survey Sampling

Following visual inspection within each 0.25 M^2 plot for surface mussels, substrate material was removed to a depth of 10-12 cm (4-5 inches) and placed onto a ¼ inch mesh screen for processing (Photos 5, 6, 7 and 8). This technique targeted greater than six-months to one-year old specimens to include large juvenile and adult stage mussels. The following information was recorded for each sample plot:

- Total width of stream at the transect location
- Number and species of mussels encountered
- Length (mm) of mussels encountered
- Substrate composition categories present (cobbles, mixed gravels, pea gravel, sand, mud, clay)

All live mussels were retained in a bucket of stream water until identified and measured, then returned to the stream unharmed. Specimens of dead and relic shells and valves were retained for identification verification and dead or relic shells that were observed along the stream bank were noted by distance from the reference point, measured, and identified when possible.

Mussel Survey Results

Water Quality

Water quality measurements in the upper survey reach for temperature, dissolved oxygen, and pH indicated typical ranges for a good quality mid-western stream demonstrating typical diurnal fluctuation. Water quality measurements taken during the mussel survey at the upper and lower survey reach reference locations are shown in Table 1. Water temperatures in the upper survey reach of West Fork Busseron Creek ranged from 28-34 °C with warmest temperature in mid-afternoon. Dissolved oxygen concentrations fluctuated with temperature saturation and in-stream algal productivity and ranged from 7.03-12.6 mg/L with highest concentrations in mid-afternoon. Specific conductivity and pH showed little temporal fluctuation and ranged from 260-296 mili-Semens/cm (µS/cm) and from 7.2-7.7 standard units (s.u.), respectively.

Water quality in the lower survey reach indicated both water temperature and dissolved oxygen concentration was influenced by the beaver dam (see Photos 13, 14, and 15). During the time of day when fluctuations of temperature and dissolved oxygen are typically near the diurnal peak, the water temperature (30.5 °C) and dissolved oxygen concentration (6.3 mg/L) were lower than observed at any time in the upper survey area (Table 1). The presence of the beaver dam did not appear to appreciably influence pH or specific conductivity. The slight increase in specific conductivity measured at the lower survey reach reference site is ecologically negligible, and may be a natural consequence of increased drainage area for this site.

Flow conditions at the time of the mussel survey reflected the dry meteorological conditions prevalent in the geographical area during several weeks prior to the survey. Flow at the upper study reach reference location was measured at 0.185 cubic feet per second (cfs), which increased to 0.323 cfs at the downstream survey reference location. The increase in flow downstream can be attributed to the contribution of local baseflow from the increased drainage area of the mine spoils within the extended riparian zone. However, it is likely the beaver dam also serves as a reservoir that contributes to maintain flow in the lower portion of West Fork Busseron Creek.

Mussel Survey

Mussel survey results indicated two mussel species, *Utterbackia imbecillis* (Paper Pondmussel) and *Ligumia subrostrata* (Pondmussel) are present in the WFBCMA (see Photos 1-4). However, all mussel specimens recorded were encountered in the upper WFBCMA study reach and no mussel specimens were found in the lower WFBCMA study reach downstream of the beaver dam.

A total 29 sample plots were surveyed in the upper WFBCMA (upstream and downstream of the reference location), which accounted for 3% of the available stream habitat within the upper study reach. Eight live and 2 recently dead or relic *U. imbecillis* specimens were found within the sample plots compared to 15 live and one relic *L. subrostarata* individual. Remains of three *U. imbecillis* mussels were found outside the sample plots on the stream bank within the study area and their original location in the stream is unknown. Extrapolations for mussel density based on the mussels encountered within the sampling frame indicates approximate densities of 1.3 *U.*

imbecillis/M^2 and 2.1 *L subrostrata*/M^2 within the study area. Table 2 provides a summary of the mussel survey results and species density and length characteristics for the upper and lower WFBCMA stream reaches.

For the lower WFBCMA mussel survey reach a total of 10 survey plots were evaluated over a 150 ft length of stream (3% of stream area) with no mussels encountered. Based on the greater number of mussels encountered in the upper study reach for an equivalent 10-frame effort and length of stream, it was determined in the field to end the mussel survey in the downstream study area and extend the study area of the upstream survey reach. While no mussels were encountered in the study plots or observed along the shore at the downstream study reach, inference from the data indicate the density of mussels in this portion of the WFBCMA is much lower than observed in the upper study reach. Because mussels were encountered at the upper WFBCMA it cannot be stated with statistical confidence that mussels are completely absent within the lower survey area. The direct and indirect effects on the presence of mussels in the lower study reach from the beaver dam and recent deposition of bed materials are unknown.

A complete listing of the location, number, size, and predominant bed material for each sample plot is presented in Table 3 for both the upper and lower mussel survey study reaches of the WFBCMA. The mussel data showed no apparent relationship or patterns that could be detected between substrate characteristics and species of mussel encountered, or evidence of spatial trends in numbers or species of mussels in the upper survey area.

Utterbackia imbecillis **Ecology**

This species is widespread in the Eastern half of the United States, with North to South ranges extending from Ontario Canada to South Texas. It is a very thin shelled, fast growing mussel species found in sandy to muddy bottomed slow creeks and ponds. Although this mussel species has been known to complete its life history without the use of fish hosts, the glochidia of this mussel are commonly found on sunfish, bullhead catfish, and large salamander larvae. This species is hermaphroditic, making it adaptable for early colonization of stream habitats. Specimens of this species in the 80 to 100mm range may be 3-5 years of age. Figure 2 shows the range in total valve length encountered for the specimens in WFBCMA.

Ligumia subrostrata **Ecology**

This species has a similar distribution to the *Utterbackia,* however the common distribution is somewhat more centered within the Mississippi River drainage. It also prefers slow to still areas with sandy bottoms, but has slightly more of preference to cleaner sand and continuously flowing creeks than *Utterbackia*. The primary host species for this mussel are sunfish. The sexes of this species are identifiable by differences in shell morphology. This species is smaller at maturity than *Utterbackia*, potentially reaching maturity at two years of age and 30-40mm in length. Figure 2 shows the range in total valve length encountered for the specimens in WFBCMA.

Discussion

The finding of two species of live freshwater mussels at the numbers encountered and the size range of specimens verifies that mussels are established in the WFBCMA. Given the relatively complex life cycle and requirements for freshwater mussels to occur in streams (see above discussion of mussel ecology) the presence of *U. imbecillis* and *L. subrostrata* in the WFBCMA is further evidence that biological function of the stream has been attained. Furthermore, mitigation of this portion of West Fork Busseron Creek was completed in 2006 and the initial sighting of freshwater mussels in the WFBCMA was in 2010, suggesting a relatively fast rate of mussel recruitment and colonization. Successful colonization of two species of freshwater mussel over a 5-year period indicates that primary biological functions of the stream were likely present shortly following remediation.

Based on literature review of growth rates for similar unionid mussels we estimate the age of the *U. imbecillis* and *L. subrostrata* specimens encountered in the WFBCMA to be up to three years in age. Breeding season and glochidia release for many mussel species is in the spring through summer seasons, so it is estimated that the specimens retained by the ¼ inch mesh screen would have been produced no sooner than spring /summer of 2010.

Tables

Table 1. Water Quality Data West Fork Busseron Creek Mitigation Area, Farmersburg IN, August 5, 2011

Site	Time (hrs)	pH (su)	Temperature (°C)	Specific Conductivity (mS/cm)	Dissolved Oxygen (mg/L)	Flow (cfs)
Upper Survey Area [1]	1100	7.23	28.5	279	7.03	
Upper Survey Area [1]	1515	7.73	34.0	270	12.6	0.185
Lower Survey Area [2]	1715	7.52	30.5	296	6.32	0.323
Upper Survey Area [1]	1820	7.62	32.9	260	11.4	

1 Upper study reach reference location at lat: 32° 14.124' N and long: -87° 21.616' W
2 Lower study reach reference location at lat: 39° 13.650' N and long: -87° 21.434' W

Table 2. Mussel Survey Data West Fork Busseron Creek Mitigation Area, Farmersburg IN, August 5, 2011

Sample No.	Location From Site Reference (ft)	Live Mussel Taxa	Relect Mussel Taxa	Total Length (mm)	Substrate Characteristics
Upper WFBC Mitigation Area Mussel Survey Stream Reach					
21	300-upstream	*Utterbackia imbecillis*		15.6	mix gravel/sand
		Legumia subrostrata		22.5	mix gravel/sand
20	285-upstream	*Utterbackia imbecillis*		46.5	mix gravel/sand
on bank	283-upstream		*Utterbackia imbecillis*	80.2	recent dead
on bank	282-upstream		*Utterbackia imbecillis*	*85.3*	fragment, approx length
19	270-upstream	*Legumia subrostrata* ♂		44.5	cobble/gravel/sand
			Utterbackia imbecillis	*41.5*	partial, approx. length
18	255-upstream	none			mix gravel/sand
17	240-upstream	none			mix gravel/sand
16	225-upstream	none			mix gravel/sand
15	210-upstream		*Utterbackia imbecillis*	79.0	valve only, recent dead
		Legumia subrostrata		17.9	cobble/gravel/sand
14	195-upstream	none			cobble/gravel/sand
13	180-upstream	*Legumia subrostrata*		26.4	cobble/clay/sand
12	165-upstream	*Legumia subrostrata*		23.8	cobble/clay/sand
11	150-upstream	none			cobble/clay/sand
10	135-upstream	*Legumia subrostrata*		25.2	cobble/gravel/clay
			Legumia subrostrata		fragment, recent dead
9	120-upstream	*Utterbackia imbecillis*		63.4	pea gravel/clay
8	105-upstream	none			pea gravel/clay
7	90-upstream	*Legumia subrostrata*		26.5	pea gravel/clay
6	75-upstream	none			mix gravel/clay/sand
5	60-upstream	none			pea gravel/clay/silts
4	45-upstream	*Legumia subrostrata* ♀		24.8	mix gravel/clay/silts
3	30-upstream	none			mix gravel/clay/silts
2	15-upstream	*Legumia subrostrata*		18.7	mix gravel/sand
1	Reference [1]	none			mix gravel/sand
1d	15-downstream	none			mix gravel/sand
2d	35-downstream	none			sand/pea gravel
3d	55-downstream	*Utterbackia imbecillis*		17.8	sand/pea gravel
4d	75-downstream	none			mix gravel/sand
5d	95-downstream	*Utterbackia imbecillis*		20.7	mix gravel/sand
		Utterbackia imbecillis		17.6	mix gravel/sand
		Legumia subrostrata		33.7	mix gravel/sand
		Legumia subrostrata ♀		32.7	mix gravel/sand
		Legumia subrostrata		29.8	mix gravel/sand
on bank	103-downstream		*Utterbackia imbecillis*	*65*	partial, approx. length
6d	115-downstream	*Utterbackia imbecillis*		23.5	gravel/sand/silts
		Utterbackia imbecillis		20.7	gravel/sand/silts

Table 2. Mussel Survey Data West Fork Busseron Creek Mitigation Area, Farmersburg IN, August 5, 2011

Sample No.	Location From Site Reference (ft)	Live Mussel Taxa	Relect Mussel Taxa	Total Length (mm)	Substrate Characteristics
		Legumia subrostrata		31.9	gravel/sand/silts
		Legumia subrostrata		29.2	gravel/sand/silts
7d	135-downstream	none			sand/clay
8d	150-downstream	none			sand/clay
Lower WFBC Mitigation Area Mussel Survey Stream Reach					
	Reference [2]				
1	5-upstream	none			mix gravel/sand/clay
2	20-upstream	none			pea gravel/sand/clay
3	35-upstream	none			pea gravel/clay/sand
4	50-upstream	none			pea gravel/clay/sand
5	65-upstream	none			pea gravel/clay/sand
6	80-upstream	none			pea gravel/clay/sand
7	95-upstream	none			pea gravel/clay/sand
8	117-upstream	none			sand/mix gravel
9	130-upstream	none			sand/mix gravel
10	150-upstream	none			sand/mix gravel

1 Upper study reach reference location at lat: 32° 14.124' N and long: -87° 21.616' W
2 Lower study reach reference location at lat: 39° 13.650' N and long: -87° 21.434' W

Table 3. Summary of Mussel Survey Results, West Busseron Creek Mitigation Area, Farmersburg IN, August 5, 2011.

UPPER WFBC MITIGATION AREA	
Total wetted stream study area (M^2)	309
Total area surveyed for mussels (M^2)	7.25
Number of 0.25 M^2 survey plots	29 (2.3%)
Total number of mussels [1]	25
Total mussel species richness	2
Extrapolated mussel density (#/M^2)	3.4
Extrapolated *Utterbackia* density (#/M^2)	1.3
Mean *Utterbackia* valve length (mm)	27.3
Range *Utterbackia* valve length (mm)	15.6 - 85.3
Extrapolated *Legumia* density (#/M^2)	2.1
Mean *Legumia* valve length (mm)	35.2
Range *Legumia* valve length (mm)	18.7 - 33.7
LOWER WFBC MITIGATION AREA	
Total wetted stream study area (M^2)	82.4
Total area surveyed for mussels (M^2)	2.5 (3%)
Number of 0.25 M^2 survey plots	10
Total number of mussels [1]	0

1 Total number of live or dead mussels within sampling frame

Figures

Figure 1. West Fork Busseron Creek Mitigation Area (WFBCMA). Stream reach reference point locations for a mussel survey conducted on August 5, 2011. Photo shown is 2008 depiction of the mitigation area and current day stream channel, but does not reflect the final and current day restoration of the immediate watershed and buffer zone.

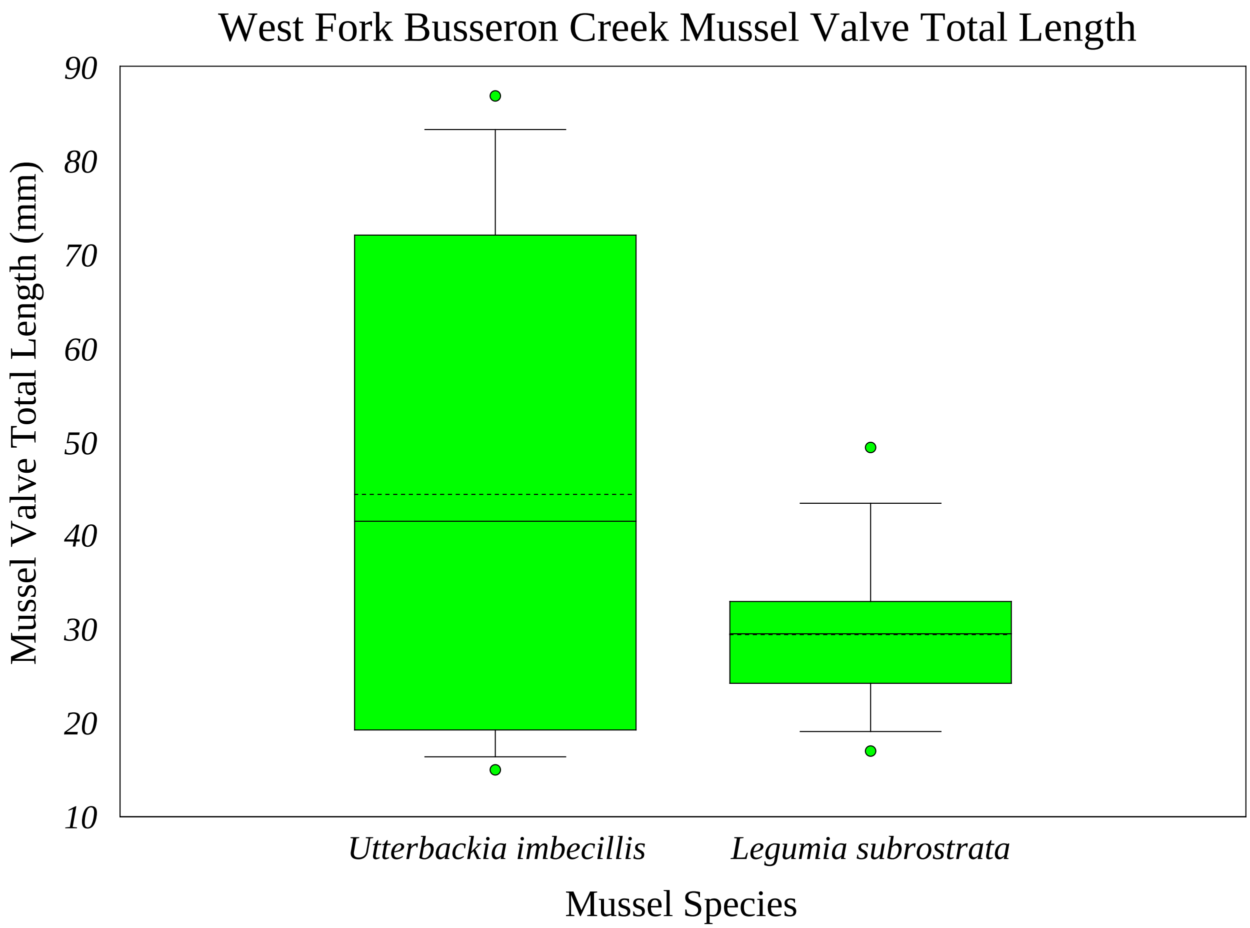

Figure 2. Box Plot of Mussel Valve Total Lengths. Values for mussels identified in survey plots in West Fork Busseron Creek, August 5, 2011. Upper and lower dots indicate 5^{th} and 95^{th} percentile range, the horizontal lines show the 90^{th} and 10^{th} percentile, the box shows the 25^{th} to 75^{th} percentile range, the dotted line indicates the mean valve length, and the solid line within the box indicates the median (50^{th} percentile) valve length.

Photos

Photo 1. Fresh *Utterbackia imbecillis* from WFBC mitigation area on ¼ inch sieve screen portion of foot mantle extruded from valves.

Photo 2. *Utterbackia imbecillis* with valve length of 63.4 mm collected from WFBC mitigation area.

Photo 3. *Legumia subrostrata* (male) from WFBC mitigation area.

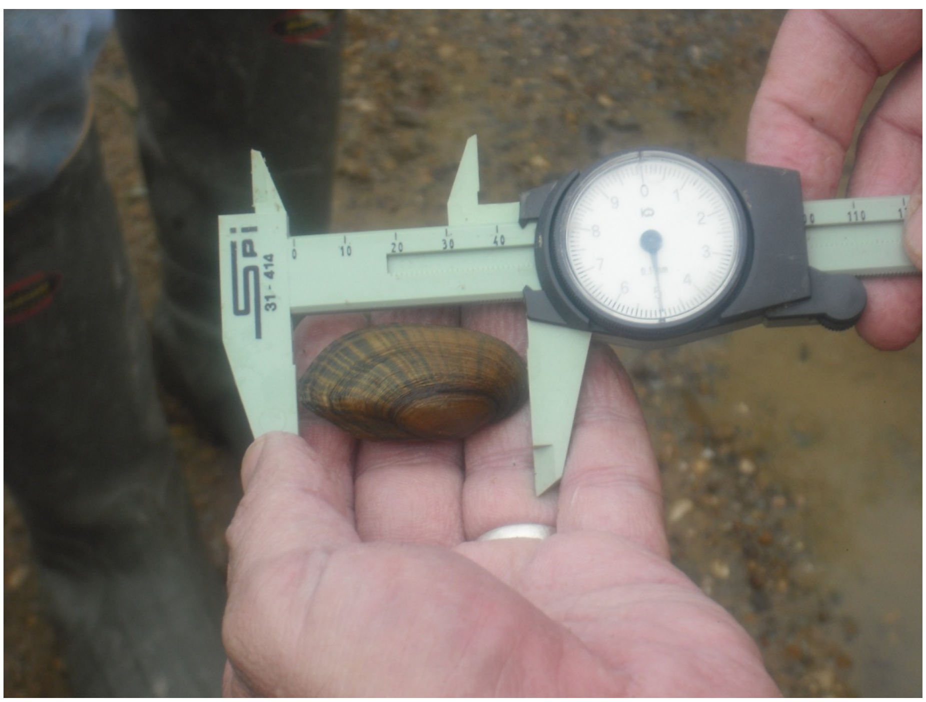

Photo 4. Ligumia subrostrata (male) with valve length of 44.5 mm from WFBC mitigation area.

Photo 5. Mussel survey 0.25 M^2 sampling frame and 0.25 in sieve screening pan.

Photo 6. Stream sediment from within mussel sampling frame being sieved in 0.25 in screening pan.

Photo 7. Field personnel checking screened substrate material for mussels.

Photo 8. *Utterbackia imbecillis* specimen from WFBC mitigation area in sieved mixed gravel on mussel screening pan.

Photo 9. Hinge view of *Utterbackia imbecillis* from WFBC mitigation area.

Photo 10. WFBC mitigation area upper mussel survey study area looking upstream from study reach reference point. August 5, 2011.

Photo 11. Typical stream conditions of WFBC mitigation area upper mussel survey study area. August 5, 2011.

Photo 12. Downstream terminus of upper WFBC mitigation area mussel survey study area, and immediately upstream of slack water due to grade control and beaver dam. August 5, 2011.

Photo 13. Downstream WFBC bioassessment mitigation area showing slack water due to presence of beaver dam. August 5, 2011.

Photo 14. WFBC mitigation area showing slack water due to presence of beaver dam. August 5, 2011.

Photo 15. WFBC mitigation area showing upstream limit of slack water due to presence of beaver dam. August 5, 2011.

Photo 16. WFBC mitigation area downstream mussel survey study area immediately downstream of beaver dam. August 5, 2011.